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LIFE SCIENCES

EFFECTS OF NONIONIZING ELECTROMAGNETIC RADIATION

No. 4

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EFFECT OF COMMERCIAL FREQUENCY ELECTROMAGNETIC FIELD ON ANDROGEN FUNCTION OF RAT TESTES

Kiev FIZIOLOGICHESKIY ZHURNAL in Russian Vol 27, No 1, Jan-Feb 81 pp 121-124

[Article by A. G. Reznikov and L. V. Tarasenko, Laboratory of Neurohormonal Regulation of Reproduction, Kiev Institute of Endocrinology and Metabolism, submitted 19 Oct 79]

[Text] Studies of the biological effects of commercial frequency electromagnetic fields (CFEMF) generated by high-voltage electric power lines are necessary in order to set hygienic standards for intensity of the electrical field. Changes were demonstrated in the reproductive system of rats and rabbits exposed for 2-4 months to CFEMF with intensity (E) of up to 15 kV/m: diminished fertilizing capacity, depressed spermatogenesis, increased excretion of 17-ketosteroids, as well as inhibition of respiration and oxidative phosphorylation in the testes [1, 2]. Impaired fertility and lower birth rate were also found in mice as a result of exposure to CFEMF with E = 50-100 kV/m [3].

As we know, the main reproductive functions in man and animals are regulated by a set of neural and humoral factors and, first of all, sex hormones. Since there is no information in the literature about the effects of CFEMF on biosynthesis and secretion of sex steroids, we studied some parameters of androgen function of the gonads of male rats exposed to CFEMF.

Methods

Experiments were conducted on mongrel albino rats weighing 250-280. The animals were "irradiated" daily for 4 months from the unit of the Kiev Scientific Research Institute of General and Communal Hygiene. Overall duration and conditions of "irradiation" with consideration of the life span of this species corresponded to the actual conditions of public exposure to CFEMF in the range of their action. The conditions for exposure to CFEMF were as follows: 1st and 2d groups, E = 15 kV/m, 20-min exposure at a time, total exposure 3 h/day; 3d group, E = 20 kV/m, 20 min, 3 h/day; 4th group, E - 15 kV/m, 80 min, 5 h/day; 5th group, E - 10 kV/m, 80 min, 5 h/day; 6th group, control for the 1st, 3d, 4th and 5th groups (first experiment); 7th group, control for the 2d group (second experiment). There were 30-min intervals between exposures. After termination of the period of exposure to CFEMF, the rats were decapitated and blood was collected in heparinized tubes. Some of the animals in the 1st and 5th groups were used in an experiment 1 month after exposure to CFEMF (these groups are referred to as 1A and 5A).

Testosterone level in blood plasma was assayed by the radioimmunological method, using the TESTOK set (International CIS, France). Radioactivity was measured with an Isocap-300 liquid scintillation counter (Nuclear Chicago, United States). We determined steroid-Δ^5-3β-ol dehydrogenase (SD) activity in fresh testicular homogenates using a modification of the spectrophotometric method based on measurement of rate of conversion of dehydroepiandrosterone into androstenedione [5]. The testes were ground in a porcelain mortar on ice; we took a batch of 115-130 mg, which was then homogenized in a medium consisting of aliquots of 0.9% NaCl solution and phosphate buffer, pH 7.4. For incubation, we prepared a mixture of the following composition: 1 mℓ 5% testicular homogenate (50 mg tissue), 2.9 mℓ phosphate buffer, pH 7.4, and 0.9% NaCl solution (1:1), 2.0 mg NAD, 100 μg dehydroepiandrosterone in 0.1 mℓ absolute ethanol. Control specimens were prepared using an analogous formula, but instead of dehydroepiandrosterone we added 0.1 mℓ absolute ethanol. The specimens were incubated in a water bath with constant stirring. Incubation at 37°C lasted 1.5 h. The reaction was stopped by putting the specimens on ice. We extracted Δ^4-3-ketosteroids from the specimens with an aliquot (5 mℓ) of purified and distilled methylene chloride. The formed emulsion was separated by centrifuging at 4000 r/min for 10 min. The extract was transferred into the cuvette of an SF-26 spectrophotometer and its optical density was measured against methylene chloride extract of the incubation medium at a wavelength of 240 nm. The increase in optical density in samples containing dehydroepiandrosterone as the SD substrate, as compared to control samples, is determined by the amount of androstenedione formed, and it serves as a measure of activity of the enzyme studied.

Androstenedione content was determined from a calibration graph of levels of extinction, which was plotted for the androstenedione standard in the range of 5-30 μg. SD activity was expressed as the amount of Δ^4-3-ketosteroids (according to androstenedione standard) in μg over a period of 90 min of incubation, scaled to the whole testis, 1 g wet tissue and 1 mg protein. Total protein content was assayed by the method of Lowry [7].

Results and Discussion

As shown by our studies, under the influence of CFEMF all of the rat testes presented a reliable decrease in size in all of the experimental groups, with the exception of the 2d, 4th and 5a (Table 1). A negligible increase in weight of the gonads was noted in the 2d group.

Testosterone content of blood plasma of experimental and control rats was characterized by significant variability within the different groups in the first experiment. Along with rather high levels (2.25-2.92 ng/mℓ), there were also rather low ones (0.33-0.72 ng/mℓ). This is related to the seasonal fluctuations of androgen activity in rat testes. The animals were sacrificed in the fall, when testosterone secretion diminishes drastically, as compared to the spring and summer period [6]. For the same reason, the mean testosterone level in control animals examined in the summer (second experiment) was 4 times higher than in the first experiment.

We failed to demonstrate reliable differences between levels of circulating testosterone in control and experimental animals, either immediately after exposure to CFEMF or 1 month after "irradiation."

Table 1. Weight of testes and testosterone content in blood plasma of rats exposed to CFEMF for 4 months (M±m)

Group No	Number of animals	Weight of testes, mg	Testosterone content, ng/mℓ
First experiment			
I	3	1286±94	1,15±0,23
		<0,05	>0,05
IA	4	1277±75	1,0±0,27
		<0,05	>0,05
III	4	1252±45	1,32±0,54
		<0,05	>0,05
IV	4	1415±111	0,89±0,23
		>0,05	>0,05
V	5	1362±58	1,23±0,35
		<0,05	>0,05
VA	3	1520±85	1,03±0,23
		>0,05	>0,05
VI	9	1640±128	0,99±0,27
Second experiment			
II	5	1578±83	2,64±0,5
		<0,05	>0,05
VII	4	1325±45	3,81±1,1

Note: Symbols > and < show values of *p* as compared to the corresponding control groups.

Table 2. Effect of CFEMF on SD activity and protein content of rat tests (M±m)

Group No	Number of animals	SD activity			Protein content, g%
		per mg protein	per gram tissue	in testes	
First experiment					
I	3	6,18±1,42	216±36	264±57	3,64±0,29
		<0,05	<0,05	<0,05	>0,05
IA	4	5,83±0,61	233±17	291±10	4,03±0,08
		<0,05	<0,05	<0,05	>0,05
III	4	6,49±1,77	194±35	266±64	3,00±0,08
		<0,05	<0,05	>0,05	<0,05
IV	4	5,18±0,56	194±26	255±42	3,63±0,12
		<0,05	<0,05	<0,05	>0,05
V	5	4,59±0,47	179±16	242±38	3,89±0,09
		<0,05	<0,05	<0,05	>0,05
VA	3	2,96±0,33	127±11	184±21	4,30±0,10
		<0,05	<0,05	<0,05	>0,05
VI	9	1,65±0,13	65±6	99±14	4,01±0,24
Second experiment					
II	5	4,68±0,17	218±16	315±32	4,67±0,33
		<0,05	>0,05	>0,05	<0,05
VII	4	2,94±0,47	175±20	225±26	5,92±0,27

Protein concentration in the testes showed a 20-25% decrease in the 2d and 3d groups of animals, and no changes were demonstrable in the other groups (Table 2),

SD activity in rat testes, like the concentration of testosterone in blood, is subject to seasonal fluctuations. The differences in enzyme activity within groups and between the two control groups were analogous to those described for testosterone concentration.

All of the animals in the experimental groups presented a substantial increase in SD activity in sex glands. In the first experiment, the degree of increase after exposure to CFEMF for 4 months was almost the same in all groups, although it was somewhat lower in the 3d group when scaled to the testis and in the 5th group when determined per 1 mg protein. In the second experiment, in spite of the fact that the absolute mean values of all estimated parameters exceeded control levels in the experimental animals, these changes were statistically confirmed only for enzyme activity per 1 mg protein. The high SD activity, as compared to the control, also persisted 1 month after termination of exposure to CFEMF. At the same time, there was a noticeable tendency toward attention of this effect, which may be indicative of the start of the recovery process.

We were impressed by the discrepancy between enhancement of enzyme activity in rat testes induced by CFEMF and absence of significant changes in level of circulating testosterone. Such a divergence could be attributed to the fact that stimulation of steroid biosynthesis does not extend to all elements of this process, as a result of which there is intensified production of Δ^4-3-ketosteroids differing structurally from testosterone. The increased excretion of 17-ketosteroids in urine of animals under the influence of CFEMF speaks in favor of this assumption. [2].

The decrease in protein concentration in the testes of some experimental groups of rats conforms with the finding of depressing effect of CFEMF on energy processes in the gonads [1].

Analysis of these data indicates that there is no correlation between changes in enzyme activity and duration, mode of CFEMF or intensity thereof in the range of 10-20 kV/m. Activation of steroidogenesis in rat testes induced by CFEMF may be interpreted as a manifestation of functional stress of sex glands as a result of involvement of neuroendocrine mechanisms in the systemic reaction.

Conclusions

1. The effect of CFEMF on endocrine function of rat testes consists of intensification of steroidogenetic process and corresponding increase in activity of steroid-Δ^5-3β-ol dehydrogenase.

2. CFEMF "irradiation" for 4 months does not elicit appreciable changes in testosterone level in peripheral blood plasma.

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FEATURES OF METABOLIC DISTURBANCES IN RAT MYOCARDIUM UNDER EFFECT OF ALTERNATING MAGNETIC FIELDS OF DIFFERENT PARAMETERS

Moscow KARDIOLOGIYA in Russian Vol 21, No 4, Apr 81 (received 20 Oct 80) pp 82-85

/Article by F. A. Kolodub, O. N. Chernysheva and G. I. Yevtushenko, Institute of Labor Hygiene and Occupational Diseases (Prof G. I. Yevtushenko, director), Khar'kov/

/Text/ The high sensitivity of the cardiovascular system, in particular of the myocardium, to the effect of constant and, especially, alternating magnetic fields was demonstrated by many authors /5, 6 and 12/. However, the biochemical mechanisms forming the basis for the development of functional and structural disturbances in the myocardium have not been clarified in many respects. N. A. Udintsev and N. V. Kanskaya /13/ have established that the effect of alternating magnetic fields leads to the activation in the heart muscle of glycolysis and glycogenolysis and to the inhibition of the pentose cycle. In the literature there are no data on the state of oxidation processes and phosphorylation associated with them, in the course of which energy needed both for synthesis and contraction processes in the myocardium is accumulated.

The study of the features of metabolism of carbohydrates, macroergic phosphates and low-molecular nitrogen compounds in the rat heart under the effect of alternating magnetic fields of various voltages and exposures was the object of this investigation.

Material and Methods

A total of 210 mongrel male rats subjected to a repeated (15 and 26 sessions) and chronic (3 and 6 months) effect of an alternating magnetic field of an industrial (50 Hz) frequency and a voltage of 32 and 7.5 kA/m respectively with a daily exposure of 5 and 3 hours in a continuous generation regime served as the objects of investigation.

Twenty hours after the irradiation of animals had been completed, the complex of indices of carbohydrate-energy and nitrogen metabolism was determined: the content of glycogen, glucose and lactic and pyruvic acid, by the method of M. I. Prokhorova and Z. N. Tupikova /7/ and the level of ATP, ADP and AMP, by the method of high-voltage electrophoresis /2/.

The intensity of oxygen consumption was assayed manometrically in Warburg's apparatus in the air atmosphere at 26°C. Succinate was used as the oxidation substrate /10/. Esterifying activity was determined according to the decrease in inorganic phosphate /11/.

The activity of glycolysis enzymes--hexokinase /1/ and lactate dehydrogenase /4/--of the pentose phosphate cycle--glucose-6-phosphate dehydrogenase /15/--of the electron transport chain--cytochrome oxidase and succinate dehydrogenase /3/--and of the oxidizing and reduced forms of nicotinamide coenzymes (NAD and NAD·H) /17/ was determined by means of spectrophotometric and fluorometric methods.

For the purpose of detecting disturbances in the nitrogen metabolism of the myocardium, the content of ammonia and glutamine /9/, as well as of amide nitrogen of protein /16/, was determined by the diffusion method.

Results of Investigation

As can be seen from the table, a repeated (15 sessions) effect of an alternating magnetic field of a voltage of 32 kA/m leads to a decrease of 20, 24 and 28 percent respectively in the content of glycogen and lactic and pyruvic acid. A small, but significant, increase in the activity of hexokinase against the background of an absence of shifts in the activity of lactate- and glucose-6-phosphate dehydrogenase is noted.

Simultaneously with the indicated changes a decline in the level of NAD·H (24 percent), in the intensity of oxygen consumption (23.7 percent) and in the esterification of inorganic phosphate (12.2 percent) is observed, while the activity of succinate dehydrogenase and cytochrome oxidase and the degree of coupling (P/O coefficient) of processes of oxidation with phosphorylation remain within the control values. The level of ATP and of its dephosphorylated derivatives also remains unchanged in experimental animals.

As can be seen from the table, of the products of nitrogen metabolism a decrease (12 percent) in the content of glutamine against the background of an absence of a change in the level of ammonia is noted.

An increase in the length of the effect of a field of a voltage of 32 kA/m to 26 sessions does not lead to an aggravation of the disturbances registered during 15 irradiation sessions with the exception of a decline (27 percent) in the level of ADP against the background of an unchanged content of ATP and AMP.

Alternating magnetic fields of a lower voltage (7.5 kA/m) lead to a rise of 41 percent in the level of glycogen in the myocardium after 0.5 months of effect, whereas after a longer period (6 months) against the background of a normalized level of glycogen a significant decrease of 18 percent in the content of glucose is noted.

Disturbances in the nitrogen metabolism of the heart of experimental rats are expressed in a significant accumulation of glutamine after 0.5 and 6 months of irradiation (38 and 22.7 percent respectively), as well as of ammonia, 3 months after the effect of alternating magnetic fields.

Indices of Metabolism and Enzymatic Activity of the Heart of Rats Subjected to a Repeated Effect of an Alternating Magnetic Field of a Voltage of 32 kA/m (Average Data of 8-12 Observation) (x±Sx)

Index	Control	Experiment	p
Glycogen, mg %	512±4.6	409±21	<0.05
Lactic acid, mg %	136.6±9.7	103.9±9.1	<0.05
Pyruvic acid, mg %	2.9±0.16	2.3±0.1	<0.01
Hexokinase, µmol of NADP·H per mg of protein per hour	0.77±0.04	0.87±0.02	<0.05
Glucose-6-phosphate dehydrogenase, µmol of NADP·H per mg of protein per hour	0.05±0.004	0.04±0.003	>0.05
Lactate dehydrogenase, µmol of pyruvic acid per g of tissue per hour	278.8±1.6	278.9±1.4	>0.05
NAD }	0.31±0.05	0.27±0.04	>0.05
NAD·H }	0.76±0.057	0.58±0.063	<0.05
ATP } µmol per g of tissue	2.75±0.3	1.92±0.2	>0.05
ADP }	1.3±0.13	1.4±0.1	>0.05
AMP }	1.3±0.17	1.0±0.08	>0.05
Succinate dehydrogenase, ΔE_{550} per mg of protein per hour	10.8±0.4	11.0±0.4	>0.05
Cytochrome oxidase, ΔE_{550} per mg of protein per hour	14.6±1.2	13.5±0.2	>0.05
Consumption of O_2, microatom of O_2 per 100 mg of tissue per hour	9.1±0.46	7.0±0.34	<0.001
Consumption of P_h, microatom of P_h per 100 mg of tissue per hour	18.4±0.8	15.6±0.7	<0.05
P/O	2.04±0.09	2.05±0.09	>0.05
Ammonia, mg %	0.92±0.02	0.9±0.04	>0.05
Glutamine, mg %	11.7±0.44	10.3±0.34	<0.05

Discussion

The data presented indicate that under the effect of alternating magnetic fields of an industrial frequency certain metabolic disturbances in the rat myocardium depending both on the voltage and on the length of effect are noted. For example, a magnetic field of a voltage of 32 kA/m after 15 sessions of effect leads to a decline in the level of glycogen in irradiated animals. Taking into consideration that simultaneously with this the activity of hexokinase increases, it can be assumed that under the field's effect an intensification in the utilization of the carbohydrates of the heart muscle occurs. Along with this the decrease in the content of lactic and pyruvic acid, as well as of the reduced form of nicotinamide coenzymes, gives reason to assume the activation under the effect of a field of the indicated voltage of the oxidizing path of carbohydrate breakdown.

The decline in the level of oxygen consumption by the heart tissue of irradiated rats, which points rather to an inhibition, not activation, of oxidation processes, is somewhat unexpected against this background. This contradiction can be explained by the possible delay under the field's effect in the oxidation of free fatty acids, which under normal conditions are the main suppliers of energy (up to 80 percent) in myocardium cells /8/.

The simultaneous decrease of a magnitude of the same order in the esterifying activity of the heart muscle of experimental animals makes it possible to conclude that the alternating magnetic field of the investigated parameters does not affect the coupling of processes of oxidation with phosphorylation.

The ability to lead to an accumulation of glycogen by the myocardium during the initial periods of irradiation, which can point to the stimulation of processes of gluconeogenesis, is the distinctive feature of the effect of an alternating magnetic field (7.5 kA/m). During a prolonged effect (6 months) a decrease in the content of glucose against the background of a normalized level of glycogen is observed. This indicates an increase in the consumption of the carbohydrates of the heart muscle during this period.

Thus, the metabolic effects of a magnetic field, in particular their direction and degree of manifestation, depend to a certain extent on the periods of the field's effect.

To the same extent the above-stated also applies to the disturbances in nitrogen metabolism expressed in the accumulation of ammonia after 3 months of the field's effect and in an increase in the content of glutamine after 0.5 and 6 months of irradiation.

Taking into account that the formation of glutamine from glutamic acid and ammonia is considered the basic mechanism of neutralization of ammonia formed during the contraction of the heart muscle [14], it can be assumed that the rise in the level of glutamine after 0.5 and 6 months of irradiation is due to the accumulation of ammonia under the field's effect during preceding periods.

Furthermore, since the accumulation of ammonium ions was observed only during individual periods of irradiation and was normalized by the end of the experiment (6 months), it is possible to draw a conclusion on the relative resistance of the metabolism of nitrogen compounds of the myocardium to the effect of alternating magnetic fields of an industrial frequency.

Conclusions

1. The direction and degree of manifestation of metabolic disturbances in the myocardium under the effect of an alternating magnetic field of an industrial frequency depend on the field's voltage and length of effect.

2. An alternating magnetic field of a voltage of 32 kA/m delays processes of oxidation and phosphorylation, not affecting their coupling, and, as compared with fatty acids, the proportion of oxidized carbohydrates increases.

3. An alternating magnetic field of a voltage of 7.5 kA/m stimulates the conversion of both carbohydrates and nitrogen substances, the degree and direction of which depend on the periods of effect.

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INFLUENCE OF ELECTROMAGNETIC WAVES IN MILLIMETER BAND ON INDUCTOPROTEIN SYNTHESIS OF PENICILLINASE BY STAPHYLOCOCCUS AUREUS

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[Article by A.Z. Smolyanskaya, A.M. Makhov, E.A. Gel'vich and M.B. Golant]

[Text] The effect of electromagnetic waves in the millimeter band on the synthesis and activity of pencillinase produced by strains of Staphylococcus aureus and Escherichia coli is examined. Experiments demonstrated that wave lengths of 6,478 and 6,468 mm inhibit the induction of the enzyme by St. aureus. However, this type of radiation does not affect the activity of the exoenzyme, released from the St. aureus strain, and also does not affect the constitutive synthesis of penicillinase by E. coli. The relationship of the effect of duration of irradiation, bacterial population and strength of current density is elucidated.

Previously, a number of authors (1,3,4) proved the possibility of a resonance effect of electromagnetic radiation in the millimeter band on biologic objects.

We examined the influence of this type of radiation on the synthesis and activity of penicillinase--an enzyme responsible for the resistance of bacteria to preparations of the penicillin group. In this report, we present results of studies which we conducted.

We selected two penicillinase-producing microorganisms: Staphylococcus aureus strain 469 with an inductoprotein type of penicillinase synthesis and Escherichia coli strain 7 with a constitutive type of synthesis of this enzyme. By means of degradation with ethylidenebromide, we established that the penicillinase gene in strain 469 is located in the plasmid.

Irradiation of the culture was conducted in Teflon plates which insignificantly absorb electromagnetic waves of the millimeter band (2). Pencillinase activity was determined mined by the method of iodometric titration (10) in a modified form (5). To study the effect of electromagnetic irradiation on penicillinase activity we used the following methodology. To a 5 ml suspension of St. aureus 469 we added a 0.05 ml solution of penicillin in a concentration of 50,000 units/ml and then 2 ml of this mixture were irradiated in Teflon plates for 1-3 hours. The relative change in penicillinase activity of the bacterial suspension was determined by comparing the activity of irradiated and nonirradiated cultures. The quantity of inactivated penicillin was measured relative to a control solution of penicillin in Hottinger's broth which did not contain a bacterial culture (introduction of killed cells in the control solution of penicillin did not exert a significant influence on the results). To study bacteria with a constitutive type of penicillinase synthesis, a bacterial suspension, inoculated according to an optical standard, was first centrifuged to eliminate the exoenzyme. The precipitate was diluted in Hottinger's broth up to the initial volume and, after supplementation with penicillin, was irradiated. As a control, we used a suspension which had not been irradiated.

To study the effect of millimeter waves on penicillinase activity in St. aureus 469, we conducted preliminary induction of penicillinase synthesis. Then, the culture was centrifuged, penicillin added to the supernatant, the culture irradiated for 1.5 hours and then the enzyme activity determined. As a control, we used nonirradiated supernatant. The magnitude of the effect was evaluated according to the relative change in penicillinase activity in irradiated and nonirradiated (control) suspensions, defined as $\eta=(A_H-A_o)/A_H$, where A_H and A_o are the penicillinase activity in nonirradiated and irradiated suspensions, respectively. In order to establish the relationship of the magnitude of effect to power current density of electromagnetic radiation, we determined penicillinase activity by a spectrophotometric method (9), which we modified. We diluted a 24-hr St. aureus culture 10 times. We used the following preparations: a phosphate buffer with a pH of 7.2-7.4, a 0.08 M solution of iodine in 3.2 M potassium iodide and a 2 percent hydrolyzate of starch. To prepare a working mixture, to a 160 ml solution of a phosphate buffer we added a 0.6 ml solution of the iodine in potassium iodide and the 40 ml hydrolyzate of starch. The final concentration of iodine in the working mixture was 240 μm.

To determine penicillinase activity, we placed 2.5 ml of the working mixture in the cuvette of the spectrophotometer (1 cm gage) and added 0.5 ml of irradiated or control cultures of St. aureus 469 with penicillin in Hottinger's broth (concentration of penicillin in culture was 2,000 units/ml). The quantity of inactivated penicillin in irradiated and nonirradiated cultures was determined relative to the solution of penicillin in Hottinger's broth in the same concentration. The bacterial suspension, given the spectrophotometric method for determination of penicillinase activity and given a significant dilution, had an insignificant effect during the course of the test on the change in optical density, not exceeding 3 percent. The amount of inactivated penicillin was measured after 20 min at 620 nm and the relative change in penicillinase activity was calculated according to the formula

$$\eta=\frac{D_{\text{о}}-D_{\text{н}}}{D_{\text{п}}-D_{\text{н}}}100\%,$$

where $D_{\text{о}}$, $D_{\text{н}}$ и $D_{\text{п}}$ are the optical density for irradiated, nonirradiated cultures and the penicillin solution, respectively.

The change in the quantitative content of protein in the phosphate buffer of St. aureus 469 subjected to electromagnetic waves was determined at 280 nm in a spectrophotometer. The 24-hr St. aureus culture was centrifuged at 6,000 rev/min for a period of 10 min, the precipitate was diluted in an equal amount of phosphate buffer and penicillin added up to a concentration of 1,000 units/ml; 2 ml of suspension was added to each plate. Some of the plates were irradiated with millimeter waves for 1.5 hours, with nonirradiated plates serving as the control. After irradiation, we took 1.5 ml of the mixture from the irradiated and from the control clutures; diluted each with 3 ml of phosphate buffer and centrifuged the sample for 20 min at 6,000 rev/min. We placed in the cuvette (1 cm) 3 ml of the liquid drawn from the irradiated and nonirradiated suspensions, determined the optical density and calculated the relative change in protein content in the phosphate buffer according to the formula

$$\eta = \frac{D_0 - D_{\text{н}}}{D_{\text{н}}} 100\%.$$

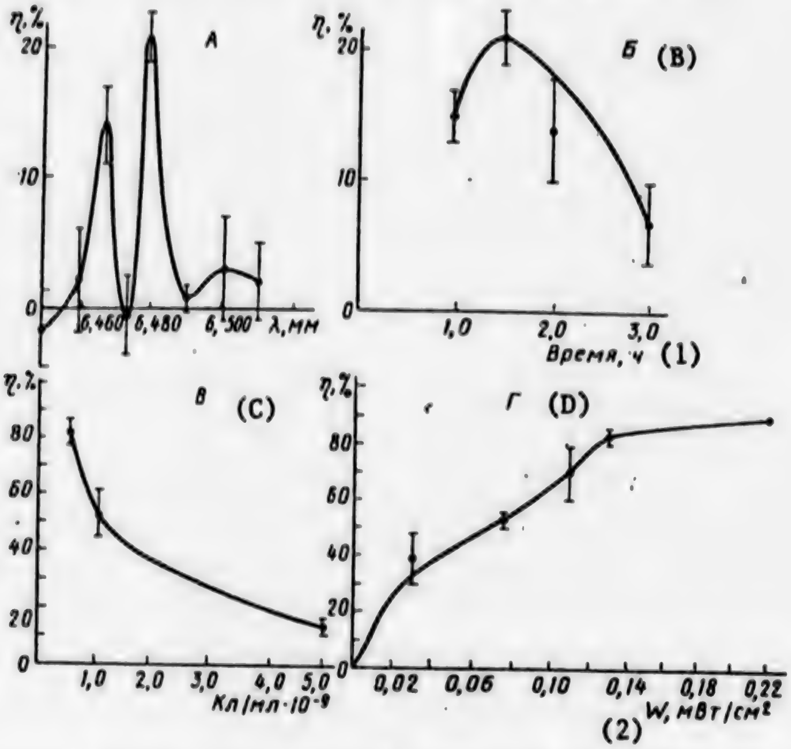
where D_O and D_H are the optical density of irradiated and nonirradiated suspensions respectively.

As the source of the electromagnetic waves of the millimeter band, we used a lamp for reciprocal waves with electronic retuning of the wave length from 5.6 to 8.0 mm. The power current density emanating from the transmitter placed at the level of the culture was decreased approximately in sequence. The value of the coefficient of the standing waves in the presence of the irradiated culture, and without it, did not differ significantly and was equal approximately to 2. The values found in the study corresponded to the power current density which reached the receiver located on a level with the irradiated cultures, in correspondence to the maximum transmitted intensity. In experiments using irradiated cultures, we also obtained correspondence to the maximum transmitted strength.

Study of the effect of elecgromagnetic waves on inductoprotein synthesis of penicillinase by St. aureus 469 was conducted using radiation with wave lengths of 6.540, 6.460, 6.468, 6.473, 6.478, 6.490, 6.500 and 6.510 mm. In preliminary tests, we established that the bacterial population in irradiated for 1-3 hours and nonirradiated cultures remained the same. However, the radiation with wave lengths of 6.478 and 6.468 mm lowered penicillinase activity. The relationship of relative change in penicillinase activity to wave length of electromagnetic radiation had a sharply resonant character. Given change in wave length to 0.005 mm, the effect disappeared. This phenomenon is clearly evident in figure, A, on which the abscissa represents wave length of the electromagnetic radiation and the ordinate represents relative change in penicillin activity of St. aureus 469. Given the effect of radiation with the active wave lengths on a culture of St. aureus, not supplemented with penicillin, accumulation or absorption of substances which influence the quantitative determination of inactivated penicillin was not detected. Cells, preliminarily killed by irradiation, bound penicillin 4% less than did nonirradiated cells. A more detailed study of resonance lines was made difficult because of the small resolution capacity of the apparatus used (0.005 mm).

The results of this study on the relationship of relative change in penicillinase activity to duration of irradiation (wave length of 6.478 mm) in room temperature conditions (22-23° C) and a suspension density of $5.0 \cdot 10^9$ cells/ml showed that maximum activity occurred during irradiation of 1.5 hours duration (figure, B). We established (see below) that the change in penicillinase activity given irradiation by electromagnetic waves was related to suppression of enzyme induction. In the nonirradiated culture, a greater amount of enzyme was accumulated and inactivation of penicillin proceeded at a greater rate than in irradiated cultures. However, with a decrease in the concentration of penicillin, the rate of the reaction in nonirradiated cultures was decreased, whereas in the irradiated culture, the rate actually increased

because of a gradual accumulation of enzyme and a small consumption of penicillin. We used irradiation of 1.5 hours duration in future experiments



Относительное изменение пенициллиназной активности Staphylococcus aureus штамм 469 в зависимости от длины волны электромагнитного излучения (*А*), времени облучения (*Б*); численности бактерий (*В*), плотности потока мощности электромагнитного излучения (*Г*). *А* — плотность суспензии $5{,}0 \cdot 10^9$ кл/мл, время облучения 1,5 ч, плотность потока мощности 0,15 мВт/см²; *Б* — длина волны 6,478 мм, плотность суспензии $5{,}0 \cdot 10^9$ кл/мл, плотность потока мощности 0,15 мВт/см²; *В* — длина волны 6,478 мм, время облучения 1,5 ч, плотность потока мощности 0,15 мВт/см²; *Г* — длина волны 6,478 мм, время облучения 1,5 ч, плотность суспензии $0{,}5 \cdot 10^9$ кл/мл

The relative change in penicillinase activity of Staphyloccus aureus strain 469 in relation to wave length of electromagnetic radiation (A), time of irradiation (B), bacterial population (C), strength of current density of electromagnetic radiation (D). A=density of suspension $5.0 \cdot 10^9$ cells/ml, time of irradiation 1.5 hours, strength of current density 0.15 mW/cm²; B=wave length of 6.478 mm, density of suspension $5.0 \cdot 10^9$ cells/ml, strength of current density 0.15 mW/cm²; C=wave length of 6.478 mm, time of irradiation 1.5 hours, strength of current density 0.15 mW/cm²; D=wave length of 6.478 mm, time of irradiation 1.5 hours, density of suspension $0.5 \cdot 10^9$ cells/ml

Key:
1. Time, hours
2. W, mW/cm²

During our study of the relationship of relative changes in penicillinase activity of St. aureus 469 to bacterial population, we found that with a decrease in the number of bacteria from $5.0 \cdot 10^9$ to $0.5 \cdot 10^9$ cells/ml, the enzyme activity increased 4 times (figure, C). Such a change has not as yet been explainable within the framework of purely physical concepts.

We studied St. aureus penicillinase activity as a function of power current density given a 1.5 hour duration of irradiation. The curve for this relationship (figure, D) shows a form of saturation, characteristic of nonthermal activity. In other words, given an increase in power density to 0.13 mW/cm^2, the impact of irradiation increases sharply, but given further increase in density to 0.22 mW/cm^2, irradiation does not have a significant impact. The significant increase in power current density can cause a thermal effect, disrupting the course of the described curve. Our data concur with results of previously conducted studies (3). The maximum inhibition of penicillinase activity, obtained in our studies, was 88 percent given a power current density of 0.22 mW/cm^2. The magnitude of the effect did not reach 100 percent, possibly because of the insufficient strength of power current density or insignificant induction of the enzyme in the irradiated culture. The cause of the incomplete suppression of penicillinase activity given an increase in its sensitivity to change in wave length of electromagnetic radiation could be the result of instability of wave length (its variation was 0.001-0.002 mm).

Studies have shown that irradiation of St. aureus 469 in a phosphate buffer by electromagnetic waves of 6.478 mm length decreased the output of exoenzyme by 12.7±1.3 percent. Given irradiation of bacterial cells without penicillin, no difference in protein content in the phosphate buffer was noted. Thus, the results obtained correspond to a decrease in content of exopenicillinase in a buffer subjected to electromagnetic radiation in relation to the entire mass of protein, released by the cells.

To resolve the question of what elements of the bacterial cells are affected by millimeter radiation (the mechanisms which synthesize the enzyme, the induction, or the enzyme itself), we conducted experiments using Escherichia coli 7 which has the constitutive type of penicillinase synthesis. Study of the effect of millimeter waves on the constitutive type of penicillinase synthesis and on the exoenzyme released by St. aureus 469 showed that radiation with these wave lengths influences neither constitutive synthesis nor the activity of the exoenzyme. We that millimeter waves disrupt the mechanism for penicillinase induction. Other authors have suggested (7) that two regulatory genes exist which control the synthesis of penicillinase by St. aureus and which function constitutively. One of the genes determines the production of the constituent part of the complex; it binds the inductor (penicillin) and induces the synthesis of penicillinase by means of inactivation of the repressor. The other gene determines the synthesis of protein analogous to the cytoplasmatic repressor and inhibits enzyme synthesis. We surmise that electromagnetic waves disrupt the mechanism for depression or binding of penicillin to the induction complex.

The second possible mechanism for electromagnetic radiation effect is inhibition of passage of penicillinase through the cell wall. Studies have shown (6) that penicillinase is contained initially in the cell wall or periplasmatic space and enters the environment through detached fragments of the cell wall.

Finally, three penicillin-binding proteins are excreted from the cell membrane of St. aureus. Membrane protein 46K has, in addition to transpeptidase, weak penicillinase activity. It is possible that this protein, not related to induction or excretion of penicillinase, also determines the insignificant inactivation of penicillin given the effect of electromagnetic waves. Seven penicillin-binding proteins are excreted from the cell membrane of E. coli (11).

Thus, the effect of electromagnetic waves of millimeter band hinders either enzyme induction or passage of penicillinase through the cell wall.

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PHYSICAL FACTORS OF ENVIRONMENT AS HYGIENE PROBLEM

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/Text/ The denaturation of the environment, which is connected with the economic activity of mankind, is not limited to chemical contamination, but is also being manifested in changes of its physical state and properties.

In accordance with the nature of the effect on the body, the physical factors can be divided into seven classes (Table 1).

The physical factors, which exist in the environment, in populated places and directly in housing, were always present in nature. However, usually the levels of the natural distinctness of many of them are relatively or even highly stable, and therefore in the past they did not attract much attention from hygienists. At present, during the age of the scientific and technical revolution, this situation is changing rapidly as a result of the progressive increase of the generation and consumption of power by mankind. Since the existing machines, mechanisms and other devices, which convert power, operate with an efficiency of less than, and often considerably less than one, some of the power consumed or generated by them in one form or another is scattered in space, increasing the distinctness of the corresponding physical factors of the environment. In this connection the urgent need is arising for the systematic detailed study of the physical factors in the habitat of man.

By the complete hygienic solution of the question we mean its elaboration in five aspects: 1) the identification, measurement and establishment of the laws of the existing state of the factors of the environment in time and space; 2) identification and establishment of the nature and degree of influence of the factors on the body; 3) the setting of standards for the factors, that is, the scientific substantiation of hygienically acceptable levels of their distinctness in the environment; 4) normalization, that is, the elaboration and introduction of technical, engineering and other decisions, which ensure the limitation of the factors at standard levels; 5) the forecasting of the hygienic situation on the basis of national economic plans and forecasts.

Table 1

Hygienic Classification of Physical Factors of the Environment

Physical factors (classes)	Decisive phenomena (factors)	Forms of manifestation (biological effects)
Mechanical (I)	Gravitation	Gravity--weightlessness
	Acceleration	Load factor
	Density of atmosphere	Atmospheric pressure
	Elastic vibrations	Firmness of support
		Noise, vibrations
Thermal (II)	Temperature of atmosphere	
	Humidity	
	Infrared radiation	Heat--cold
	Mobility of the air	
Optical (III)	Radiation of light	Illumination
	Ultraviolet radiation	Color spectrum
		Indirect
Electrical (IV)	Electrostatic field	Indirect
	Electric current	Indirect
	Ionization of atmosphere	Indirect
Magnetic (V)	Magnetic field	Indirect
	"Magnetization" of atmosphere	Indirect
Electromagnetic (VI)	Electromagnetic field of various radio frequency bands	Indirect
Ionizing (VII)	X-ray radiation	Indirect
	Nuclear radiations	Indirect
	Cosmic radiations	Indirect

If we look at the hygienic elaboration of the problem of the physical factors of the environment from this standpoint, considerable irregularity is found (Table 2).

The thermal and optical factors, as well as noise are the most studied both in the general hygienic respect and from the standpoint of environmental hygiene. Such mechanical factors as acceleration--the load factors in transportation, the firmness--elasticity of support, vibrations, as well as many aspects of the problem of the electrical, magnetic and electromagnetic factors have been elaborated to a lesser extent, especially in the latter respect.

But if we take into account the fact that until relatively recently the physical factors had practically not been studied by hygienists and the bulk of the corresponding research was performed in recent years precisely in our country, it is impossible not to see certain achievements of Soviet hygiene in the elaboration of this problem. The scientific and organizational activity of the Section of Physical Factors of the Union Problem Commission "The Scientific Principles of Environmental Hygiene," which was established several years ago, unquestionably is promoting this. The indicated section is stepping up the elaboration of the methodology of setting hygienic standards of physical factors, is organizing the preparation of comprehensive programs on the problem, approves the results of research, examines the materials on standards of hygiene and makes the appropriate suggestions for approval by the USSR Ministry of Health.

Table 2

Hygienic Elaboration of the Problem of Physical Factors of the Environment

Class	Factors	Aspects of scientific elaboration					
			nature and degree of influence				
		degree and laws of distinctness	on the body	on the environment	setting of standards	normalization	forecasting
I	Gravitation	+++	+	-	-	-	+++
	Acceleration	±	±	-	-	-	+++
	Atmospheric pressure	+++	++	+	+	+	++
	Firmness of support	±	-	-	-	-	+
	Noise	+++	+++	-	+	+	++
	Vibrations	±	-	-	±	±	±
II	Temperature	+++	+++	++	++	++	++
	Humidity	+++	++	++	++	++	++
	Mobility of the air	+++	++	++	++	++	++
	Infrared radiation	++	++	+	+	+	+
III	Illumination	+++	++	+	++	++	++
	Color spectrum	±	±	-	-	-	+
	Ultraviolet radiation	++	++	+	++	+	+
IV	Electrostatic field	±	+	-	+	±	-
	Electric current	-	±	-	-	-	-
	Ionization of the air	++	++	+	+	+	-
V	Magnetic field	±	±	±	-	-	-
VI	Electromagnetic fields of various radio frequency bands	+	++	-	+	+	+
VII	Ionizing radiations	++	+++	++	++	+	++

One of the relatively new and important directions of the scientific research in the hygiene of physical factors is the determination of the nature, degree and main laws of the biological action and the hygienic significance, as well as the setting of standards in populated areas and everyday life of the electrical and electromagnetic properties of the environment.

Although electricity as a physical phenomenon exists regardless of atmospheric air, under the real conditions of the atmosphere of the earth all the basic electrical factors (the electrostatic field and electric current in the atmosphere) in one way or another are connected with the ionization of the air (aeroionization), since it determines the other electrical properties--the nature and size of the space charge and the electric conductivity of the air.

Following the fundamental works of L. L. Vasil'yev (1953), A. A. Minkh (1963) and numerous works of other authors it is not to be doubted that aeroionization does make a difference for the human body. It has been shown that moderately increased ionization of the air (up to 2×10^3 to 3×10^3 light ions per cm^3) has a beneficial, normalizing effect on the body (F. G. Portnov, 1972, 1976, and others), while higher concentrations of aeroions (more than $5 \times 10^4/cm^3$) leads to adverse biological effects (M. G. Shandala, 1974). At the same time we have been able to show that not only the overall concentration, but also the mobility of the ions (light, heavy), the amount of the absolute predominance of ions of one sign of charge or another and the dynamicity of aeroionic conditions are of importance.

With allowance made for everything that has been mentioned, the plenum of the Section of Physical Factors of the Union Problem Commission "The Scientific Principles of Environmental Hygiene" recommended that the USSR Ministry of Health fix the aeroionic conditions of residential, public and production premises, having set the maximum permissible concentrations at the level of 5×10^4 positive and negative light ions per cm^3 of air with an absolute predominance of not more than 5×10^3 under the conditions of dynamic aeroionic state and by a factor of 10 less (5×10^3 and 5×10^2 respectively) with a stable state of aeroionization.

The rapid growth of production, which has been observed in recent decades, and the extensive use of electrifiable polymeric materials in construction, in the manufacturing of furniture, clothing, footwear, other everyday items and so forth have led to a sharp increase of the distinctness of static electrification and static electric fields in the environment. In this connection considerable contingents of people are being drawn into the sphere of influence of static electric fields.

The works of a number of authors have shown that a static electric field is a biologically active factor of the environment (F. G. Portnov, 1972; K. I. Stankevich et al., 1972; V. Ya. Akimenko, 1974). Moreover, the primary effects of the influence of a static electric field on the human body are manifested in the decrease of sensitivity to some adequate stimulations (tactile, temperature, pain), the decrease of the flow of blood in the skin, the decline of the tonicity and reactivity of the sympathetic section of the autonomic nervous system, the depression of oxidation-reduction processes in the surface layers of the skin and others. It might be thought that integuments are a unique triggering mechanism in the development of the effects of the influence of static electric fields on the human body.

The generalization of the materials gathered in the country, which was carried out on the instructions of the Section of Physical Factors, made it possible to substantiate scientifically the threshold of the chronic effect of static electric fields at the level of 30 kV/m and to recommend that the USSR Ministry of Health approve as the maximum permissible level of static electric fields 20 kV/m.

In recent decades in connection with the rapid and universal development of radio broadcasting, television, radar, electric power transmission and so forth the intensity of artifical electromagnetic fields of various frequency bands has increased sharply in the environment. As a result, under the conditions of modern populated areas the indicated factor is acquiring an increasing hygienic importance.

The biological activeness of high levels of electromagnetic fields and their occupational health significance, which is connected with this, have been known for a comparatively long time. As to the effects of electromagnetic fields of low intensities, which are being detected in the environment and daily life, that is, the communal hygiene and general hygiene aspects, they were the subject of active study over the past 10-15 years (Yu. D. Dumanskiy et al., 1975; M. G. Shandala, 1979). The nature and degree of the biological effect of electromagnetic fields depend on many factors, among which the properties of the radiation itself, first of all the frequency, are of primary importance. In addition to the previously studied shortwave and ultrashortwave bands in recent years the nature, degree and main laws of the biological effect of electromagnetic fields of superhigh and low frequencies and medium-wave radio emissions have been studied.

Superhigh frequencies or microwaves are the most biologically active frequency band of electromagnetic fields. By means of physiological, immunological, pathomorphological, histochemical, as well as psychological and other methods of research the biological activeness of not only thermal levels of the field of superhigh frequencies (more than 1 mW/cm^2), but also of levels which cause a detectable thermal effect, was shown by Soviet scientists and has now been confirmed in other countries. At the same time the nature of the biological reactions did not reveal any appreciable specificity: changes on the part of the nervous and cardiovascular systems, metabolic processes and immunological reactivity, which are accompanied by some histomorphological changes, are found. A distinctive feature of the biological effects of nonthermal levels of superhigh frequencies is their reversibility. This concerns the observable decrease of the activity of the central nervous system, symptoms of fatigue and the feeling of tiredness, vagotonic and dystrophic changes of the cardiovascular system and others.

The recognition of the hygienic significance of electromagnetic fields of an industrial frequency of 50-60 Hz is an event of the present decade. In the USSR the interest in electromagnetic fields of industrial frequency as a possible harmful factor appeared in connection with the development of high voltage electric power transmission lines and has increased considerably in recent times at the same time as the extension of their networks and the increase of the voltage in them.

The analysis of the results of experimental and full-scale studies, particularly those conducted at the Kiev Institute of General and Communal Hygiene imeni A. N. Marzeyev, shows that the long-term systematic exposure to electromagnetic fields of industrial frequency with an intensity of more than 1,000 V/m has an influence on the body, which is manifested in changes of the functional condition of the nervous, endocrine (the activation of the hypophysis-adrenal cortex system and the slowing of the function of the thyroid gland), cardiovascular and generative systems, the change of metabolic processes and others. The indicated changes can be regarded as a significant straining of the compensating mechanisms in response to the effect of electromagnetic fields of industrial frequency. Extensive experimental studies on animals and observations on people have been made in this respect (A. A. Minkh et al., 1975). The nature of the propagation of an electromagnetic field, which is created by radio stations of the middle band, radar stations and high voltage electric power transmission lines both in open areas and on the territory of residential development, was studied at the same time. The first studies were made of the state of health of the population in connection with the effect of electromagnetic fields (A. M. Serdyuk, 1977), as well as of the compensatory-adaptive reactions of the body under the influence of the energy of superhigh frequencies of low intensity (M. G. Shandala and M. I. Rudnev, 1975). The results of all these studies made it possible to establish the laws of the propagation of electromagnetic fields under the conditions of populated areas, to determine the significance of the biological effects which are detected under the influence of this factor and to substantiate scientifically the maximum permissible levels for the population of electromagnetic fields of industrial frequency, the middle band and superhigh frequency.

On the basis of previous, as well as new data "The Health Norms and Regulations" for the location of radio and television broadcasting and radar facilities (Construction Norms and Regulations No 1823-78) were elaborated at the Kiev Institute of General and Communal Hygiene imeni A. N. Marzeyev and were approved in 1978 by the USSR Ministry of Health.

In addition to questions of the health appraisal of electromagnetic fields as an environmental factor, works completed in recent years have outlined ways of solving the problem of protecting the population from the adverse effect of electromagnetic fields (S. I. Dumanskaya, 1973). Various organizational and technical, public health, design, construction, preventive medical and other measures, the comprehensive utilization of which is making it possible to find the optimum solutions in different situations, have been proposed for this purpose.

Another important direction of scientific research in the hygiene of physical factors is the study of the biological action, the determination of the hygienic significance and the setting of standards in populated areas and daily life of the mechanical properties of the environment (noise of different qualities, subsonics and low frequency vibration, the acceleration of transportation, the firmness of support--the coverings of the floor, furniture and others). A significant number of works of an applied scientific nature, which touch upon questions of noise emission, have been completed in recent years on the level of these scientific directions. They are works on the plotting of maps of the noise of large ci (I. A. Shishkin et al., 1972; I. P. Los' et al., 1972; V. A. Tokarev et al., 1972). Their goal was the study of the acoustic conditions on the main transportation routes of cities with allowance made for the traffic density and various design decisions, the hygiene assessment of the levels of noise, which are registered in the residential housing system, and the elaboration of recommendations on the controlling of this factor. The conducting of these studies originated from the needs of the practical organs of public health, which need for everyday use materials which make it possible to evaluate urban development situations (from the point of view of acoustics) at the stage of the allotment of parcels of land and the planning assignment. The preparation of "Procedural Recommendations on the Plotting of Noise Maps" was the scientific result of the performed work.

A forecast of the acoustic conditions of the studied cities for the future, which covers the period to 1990-2000, was compiled at the same time as the evaluation of the present state (Zh. I. Chernaya et al., 1975).

The study of the physiological influence of noise on the body is being carried out in the direction of the establishment of the nature and main laws of its biological action given various energy and time characteristics, as well as the solution of questions of the combined and united effect of noise with other factors of the habitat. The studies of the first direction made it possible to elaborate drafts of health standards of noise, including the noise created by rail and air transport, and to make more precise the health protection zones of separation between the residential housing system and the corresponding projects (I. L. Karagodina et al., 1972; L. A. Oleshkevich et al., 1972).

The nature and main laws of the biological effect of noise should be assessed with allowance made for its energy and time characteristics. A number of studies, which have been performed in this direction on the hygiene study of intermittent and pulsed noises, which are characteristic of urban transport, made it possible to establish that the degree of influence of isoequivalent noises on the body depends on the time characteristics of the influencing noises. It was shown that noises of the identical equivalent level and spectral composition, but of a different discontinuity are not analogous factors of influence on the body. An identical effect of the influence of noises can be observed in the case of a difference of the maximum levels of up to 10 dB (L. A. Oleshkevich and I. I. Karachev, 1975). The completion of these studies will promote the setting of sounder hygienic standards of noise with allowance made for both its energy and its time characteristics. A number of studies have as a goal the establishment of the influence of noise on the ability to do mental work, the study of the influence of urban noise on patients, particularly patients with hypertensive disease and neuroses (V. I. Pal'gov and Zh. I. Chernaya, 1972; V. I. Pal'gov and S. I. Eppel', 1970).

A study was made of the noise conditions in apartment houses and public buildings, which are created not only by penetrating transportation noise, but also by internal sources (the engineering, technological and sanitation equipment, household appliances). These studies were aimed at the elaboration of hygienically sound norms of the sound-proofing of protecting structures (G. L. Osipov, 1972).

In order to optimize the health norms with allowance made for the noise load received by a person, the Moscow Institute of Hygiene imeni F. F. Erisman conducted a survey of the urban population living on thoroughfares with heavy transportation traffic. Here particular attention was devoted to the detection of the occurrence of hypertension and ischemic disease among the residents of the noisy regions of large cities.

The questions of the development of pathology and the state of health of the population are closely connected with the problem of adaptation to urban noise. An independent work was devoted to the study of the interaction of these processes. Studies of the adult and child population, as well as experiments on animals are being conducted.

The generalization of the numerous studies of urban noise, which have been conducted in different cities of the Soviet Union, made it possible to make a national forecast on the theme "Communal Hygiene Aspects of Noise," which became a part of the international forecast on this theme, which is being made within the framework of cooperation with the CEMA member countries.

Along with noise, various technical devices within the home, as well as urban transport can generate in residential and public buildings such undesirable mechanical oscillation as vibration. So far the few studies, primarily by foreign authors, show that even a small vibration, but one perceivable by a person, in housing is always perceived as an alien and disturbing factor, which has, like noise, not only an unfavorable physiological, but also psychological influence (Schplittgerber).

In recent years in our country standard levels of vibration, which are based on the recommendations of the International Organization for Standardization, as well as on some domestic studies (I. G. Kuz'michev and M. I. Shelukhin, 1971; V. V. Shishkina et al., 1972), were proposed and in 1975 were approved by the USSR Ministry of Health (Construction Norms No 1304-75). But this does not at all eliminate the question of the need for the further thorough study of the nature and laws of the biological effect of this factor, since the materials of a survey of the population, that is, subjective evaluations of the influence of vibration of various parameters on the body, were the basis for the adopted standard. In this connection the indicated standards should be regarded as temporary and subject to thorough scientific substantiation.

Acceleration, to the influence of which considerable contingents of the population are regularly subjected both in urban transport and when using elevators, is also a biologically active factor. As preliminary studies have shown, the ascent and descent in an express elevator are accompanied by hemodynamic changes and subjective disturbances, especially among people of retirement age. However, the hygienic importance of this factor for the present has been inadequately studied, in connection with which the corresponding scientifically sound regulations are absent.

Apparently, the problem of accelerations in urban transport is and especially will be in the future of even greater importance. With the existing average traffic speeds in the city of up to 20 km/hr passengers experience accelerations (positive and negative) from 2-3 to 48 times per minute, on the average of 0.86-1.44 m/sec^2 with vibrations from 0.3 to 3.5 m/sec^2. The duration of the effect of acceleration is even more variable--from 0.2 to 30 sec (K. D. Feshchenko, 1975).

On the basis of the notions that rectilinear accelerations cause stimulation of the otolithic apparatus and the cumulation of such stimulations leads to the development of "motion sickness" syndrome, it may be thought that under the conditions of the daily repeated use of passenger transport the effect of acceleration is not an indifferent factor.

Thus, unquestionable gains have been made in our country in the scientific elaboration of the problem of the hygiene of physical factors of the environment. However, as follows from the above, many questions of this important and relatively new problem still require solution. In this connection, as well as on the basis of forecasts of the substantial increase of the distinctness of various physical factors the stepping up of the efforts of hygienists and other specialists in the most important scientific directions of the problem is necessary. Precisely for this reason a number of questions of the latter are the subject of scientific cooperation of the USSR with a number of socialist CEMA member countries, as well as with the United States.

The ascertainment of the hygienic significance of combinations of various physical factors, as well as of their combined effect with chemical contaminants and biological agents also seems very important. Such complexes are real for the conditions of many modern populated areas, and their experimental study should be the necessary basis for the launching of the most important operations at the present stage of public health on the study of the health of the population in connection with the combined influence of natural and anthropogenic factors of the environment.

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EFFECTS ON BIOLOGICAL SYSTEMS OF ELECTROMAGNETIC OSCILLATIONS IN MILLIMETER RANGE OF WAVELENGTHS

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[Article by N. D. Devyatkov, O. V. Betskiy, E. A. Gel'vich, M. B. Golant, A. M. Makhov, T. B. Rebrova, L. A. Sevast'yanova and A. Z. Smolyanskaya, Institute of Radio Engineering and Electronics, USSR Academy of Sciences, Moscow, submitted 14 Jun 79]

[Text] The main patterns of effects of millimeter radiation of nonthermal intensity on various biological objects are discussed on the molecular, cellular levels, as well as on the level of complex living organisms. The main objects of the studies were hemoglobin and erythrocytes of human blood, E. coli, Staphylococcus aureus, Aspergilla mold, mice and rats.

Studies of the effects on biological objects of electromagnetic oscillations over a broad range of wavelengths, from meter to submillimeter (nonionizing radiation) ranges, have been in progress for about 50 years. When analyzing this problem, following the existing tradition the researchers paid attention primarily to the thermal effects of interaction [1]. Millimeter (mm) and submillimeter (submm) radiations occupy an intermediate place between the radio-frequency and optical ranges; for this reason, both the wave and quantum approaches are equally effective for describing the phenomena induced by these waves. It is logical to assume that the distinction of this frequency range could also be manifested in the interaction of radiation with a substance.

The thought that electromagnetic oscillations in the mm range of wavelengths could have a specific effect on biological structures and organisms was voiced in the USSR in about 1964-1965 [2]. By that time, generators of coherent oscillations covering this frequency range had been determined, and this provided the technical prerequisites for the first scientific investigations. The hypothesis was expounded that living organisms could not have natural adaptive mechanisms for oscillations of appreciable intensity in this range, since mm radiation of extraterrestrial origin is absorbed almost entirely by earth's atmosphere. Of greatest scientific and practical interest was the effect of small, nonthermal doses of radiation, of the order of several mW/cm^2, on various objects for the purpose of selective change in properties or functions of the irradiated object. True, many researchers believed that work in this direction may hold no promise, since the quantum of energy is very small in this range and constitutes 10^{-4}-10^{-3} eV, which is less not only than the energy of such weak interactions as hydrogen bonds (10^{-1}-10^{-2} eV), but of thermal motion (kT [?] ~$2.5 \cdot 10^{-2}$ eV). However, the possibility of such an effect is attributable to the fact that mm radiation has quanta of energy that correspond in order of magnitude to excitation of various macromolecular or cellular structures, as well as the closeness of frequency of electromagnetic oscillations in this range to the intrinsic frequencies of rotation of macromolecules or molecular groups (particularly in cases where the molecule or individual molecular groups of complex molecules have a dipole electric moment), and the possibility of changing, for example, the conformational state of molecules. One could have then expected resonance effects. The intensive experimental research (some of the results of which are described below) that was deployed confirmed the validity and fruitfulness of studies in this direction. There are rather comprehensive and well-argumented Soviet publications on the effects of mm radiation on various biological and organic objects [2-10]. Only recently has intensive research begun abroad on the specific effect of radiation on biological structures and organisms differing in complexity [11-16]. As for the effects of mm radiation in high, thermal doses, the mm range is unlikely to differ in any way from other ranges of electromagnetic oscillations, with the exception of depth of penetration of radiation, which is of the order of hundreds of microns.

In the following, we shall discuss the results of some studies conducted in our country on the effects of mm radiation on various biological objects on the molecular, cellular levels and on the level of complex living organisms. It must be noted that mm radiation can also be used well for the study of intermolecular interactions, as well as to determine the role of water in the function of biologically active compounds. Original results of spectroscopic studies have been reported [17-21].

Distinctions of Effects of Millimeter Radiation of Nonthermal Intensity on Biological Objects

First of all, we must mention the resonance nature of these effects. The frequency band in which they are effective usually constitutes 0.01-0.001 of mean frequency. Beyond this narrow frequency band, there is no effect. Not infrequently, one observes several frequency bands, in which exposure is effective, and sometimes the effect may present a different sign depending on frequency of oscillations, for example, the result of exposure may have a stimulating or depressing effect on the biological object. The existence of alternating narrow resonance bands, in which the effect of exposure is manifested, was the main obstacle to reproduction of experimental results at the early stages of development of mm radiation generators and measuring equipment, when it was impossible to provide sufficient stability of radiation frequency.

Another unusual feature is that the effect of exposure depends little on radiation power flux density. If power flux density exceeds a certain threshold level, a change in oscillation power by a factor of 10^2-10^5 in the range of nonthermal intensity does not alter the effect of exposure. Occasionally, even if an object starts to warm up, the effect still remains unchanged, which indicates that thermal heating is not the basic factor in manifestation of the effect. Thus, the concept of energy dose, which is widely used with reference to ionizing radiation, loses its meaning here.

The influence of time of irradiation on the effect is also distinctive. In the case of brief exposure (~15 min) the effect is often virtually undemonstrable. With increase in irradiation time, the effect of exposure appears and its effectiveness increases; after this, one gradually observes saturation, i.e., the magnitude of the effect reaches a certain maximum and holds at this level with increase in irradiation time. Occasionally, after reaching a maximum the effect of exposure may diminish. In some cases, to obtain a significant effect multiple exposure to radiation, up to 10-20 times, was used at certain intervals. For microorganisms this corresponded to irradiation of many generations. In some cases, even a negligible effect began to be demonstrable only after several irradiation procedures.

Nor can we fail to mention the influence of the stage of biological development on the effect of exposure, which was observed both in our country and abroad. Exposure to electromagnetic waves can lead to depression, stimulation of some function in a living organism, or have no effect, depending on the phase of development. Obviously, this function can be observed in pure form only in organisms with a very long (lasting many days) cycle of biological development.

Finally, the strict reproducibility of the results of exposure is another of the observed typical distinctions of the effects of mm waves on biological objects. If the main distinctions of interaction between radiation and biological structures studied are taken into consideration in setting up the experiments, the scatter of experimental data from the tests is much smaller than the magnitude of the observed change. Statistical processing of series of experiments is often virtually unnecessary for demonstration of the result or evaluation of magnitude of effect. This is a very important circumstance, since the difficulty of setting up and conducting studies makes it difficult to repeat experiments for, for example, hundreds of times, which is important to reliability of statistical processing when there is a wide scatter of experimental results. Moreover, the high reproducibility of the results is an important distinction of the effects of nonionizing radiation, as compared to ionizing radiation, when the quantum energy is sufficient to destroy any structural element, and for this reason the result of exposure is a random element, which is governed by statistical rules.

Objects Exposed and Results of Experiments

As we have already noted above, interaction between electromagnetic oscillations in the mm range with biological structures cannot (by virtue of low quantum energy) lead to a break in molecular bonds. But it can lead to changes in configuration and affect biochemical processes in these structures or the nature of their interaction with ionizing radiation. We describe below some of the results of studies conducted on the molecular, cellular and organism levels.

The existence of nonthermal (resonance) effect of millimeter radiation on hemoglobin (Hb) in a lyophilized state and in aqueous solutions irradiated in the wavelength range of $\lambda = 6-8$ mm has been demonstrated in several works using chemical methods of analysis [22], as well as Mossbauer spectroscopy [23]. Two mutually complementary hypotheses have been expounded on the mechanisms of the effect of radiation on Hb: 1) the effect could be related to the influence of radiation on the molecular group in the active center of Hb, distal histidine E7, which may effect rotatory rocking at a frequency in the mm range and affect the nature of the bond between heme and globin; 2) mm radiation may affect the Hb molecule as a whole and excite resilient oscillations of the entire protein body, also in the mm range, and this excitation may also be transmitted to histidine E7 ("protein machine" hypothesis [24, 25]). It was found that in aqueous Hb solutions the sign [direction] of the effect of radiation (degree of heme-globin bond) may vary, depending on the wavelength of electromagnetic oscillations. This was demonstrated very graphically upon irradiation of aqueous solutions in the wavelength range of λ = 1-2 mm [17]. Below are the experimental results averaged for 10 experiments:

λ, mm	0.9	1.13	1.58
$\eta \pm \sqrt{\sigma^2_\eta}$	1.1+0.03	0.83+0.08	1.17+0.11

Here, η is the mean ratio of depth of heme-globin dissociation in the experiment to depth of dissociation [splitting] in the control, $\sqrt{\sigma^2_\eta}$ is the mean square deviation. We see that radiation can have both a stabilizing and destabilizing effect on the heme-globin bond in the Hb molecule. If we consider that with λ~2 mm absorption in aqueous solutions constitutes about 40 dB/mm, apparently transmission of the effect over the entire irradiated solution, with layer thickness of 1 mm, is possible, for example, due to diffusion.

Studies were also made of the effects on stability of erythrocyte membranes of mm radiation in the ranges of λ~1-2 and 6-8 mm [26]. It was found that, with λ = 1.15, 1.58, 1.73, 6.25, 6.4, 6.5 and 6.73 mm, radiation had a predominantly destabilizing effect on erythrocyte membranes, and an appreciable effect was observed with 60-fold or greater dilution of erythrocytes.

Most studies were referable to the effects on microorganisms and animals. We list briefly below the results of some of these studies. We shall start with examples of studies made of microorganisms, in which the effect of exposure was demonstrable after single irradiation. Thus, in studies of the effects of electromagnetic oscillations in the mm range on E. coli C600, colicin synthesis was chosen as the test [27]. The effect was characterized by the induction coefficient, i.e., ratio of percentage of microorganisms producing colicin in the irradiated culture to the percentage of microorganisms producing colicin in a nonirradiated culture. All other conditions (except radiation) were the same in the control and for the irradiated object. The frequency bands, each of the order of $\Delta f/f \sim 10^{-3}$, in which the coefficient of induction reached 2-3, were in the area of 5.8, 6.5 and 7.1 mm. Saturation of the effect was observed with power flux density in excess of 5 μW/cm^2. Exposure time was at least 30 min, and optimum time was 2-3 h. Let us mention that investigators relate induction of colicin synthesis to the effect on the episome. Thus, one can relate the effect of electromagnetic radiation on colicin production to the influence on the cell's extrachromosomal genetic system.

A study was also made of the effect of electromagnetic radiation on induction of penicillinase synthesis by Staphylococcus aureus (Figure 1). The amount of inactivated penicillin in the irradiated culture, determined by the spectrophotometric method, served as a criterion of effectiveness of irradiation. The wavelength, at which an effect was observed, was in the region of 6.48 mm. The band width of frequencies, with which an effect was observed, constituted $\Delta f/f \sim 10^{-3}$. Power flux density corresponding to saturation of the radiation effect constituted ~0.1 mW/cm^2. Optimum exposure time was ~1.5 h. The effect increased linearly with dilution of the bacterial suspension. The change in penicillinase synthesis reached 80% with irradiation.

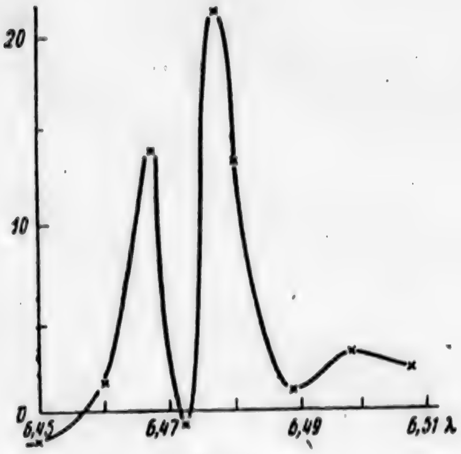

Figure 1.
Relative changes in penicillinase synthesis in staphylococcus cells as a function of wavelength, $5.0 \cdot 10^9$ cells/mℓ; irradiation time 1.5 h; power density 0.15 mW/cm^2. X-axis, wavelength, mm; y-axis, amount of inactivated penicillin:

$$\frac{A_n - A_0}{A_n} \cdot 100\%$$

Here are some examples of experimental work, in which an appreciable effect of radiation was observed only after numerous exposures of microbiological objects [28]. For this purpose, a study was made of the effect of repeated exposure to electromagnetic radiation on Aspergillus mold (Aspergillus oryzal), which synthesizes proteases that effect hydrolysis of human blood fibrin. The practical implications of this study are referable to development of products to be used for the treatment of thrombosis. A sporulated suspension of the mold in water was exposed to radiation 10 times. At wavelengths of 6.4-6.5 mm, there was 2-2.5-fold increase in fibrinolytic activity. This is a significant result, since it is difficult to obtain mutant forms of microorganisms that would increase synthesis of enzymes with fibrinolytic action using other chemical and physical factors. Moreover, with such factors, the mutant specimens were severely damaged, they developed poorly and required enriched media for their development. Specimens that appeared in the case of exposure to the mm range of waves developed well on ordinary media and, as we have already mentioned, had increased activity [28].

There was another work dealing with the effects of electromagnetic waves in the mm range on the yeast-like fungus, Endomycopsis (End. filulger), which is used as an amylolytic enzyme producer in the alcohol industry for hydrolysis of raw material containing starch, with production of glucose.* In this case too, repeated (7 times) exposure to radiation at wavelengths of 6.0 and 6.2 mm of the fungus increased the amylolytic activity of the culture by 50%. The culture retained increased activity for about 1 year.

Finally, we shall mention experiments on the effects of electromagnetic waves in the mm range on Chinese silkworm eggs.** These studies were conducted in order to demonstrate the influence of the phase of biological development at the time of irradiation on the effect of the latter. The eggs were synchronized and started to develop simultaneously. The time of development to caterpillars is about 2 weeks. Accordingly the material was divided into several parts, with 1100-1200 eggs in each. Each part was irraited once for 3 h in the course of the developmental period, and each part was irradiated on some day of the developmental cycle. They were irradiated before appearance of caterpillars. Irradiation conditions were as follows: λ = 6.48 mm, power flux density 10 mW/cm^2, the object was in the open space between the transmitting and receiving funnel [horn] antennas. During exposure, the transmitted power and SHF wavelength were controlled [monitored]. The results of exposure were evaluated on the basis of percentage of eggs from which caterpillars developed. Eggs exposed within the first 4 days yielded the same percentage of caterpillars as control eggs. Irradiation on the next 4 days led to a 2.5-fold decrease in percentage of hatched caterpillars. Finally, irradiation on the last 4 days increased by about 1.5 times the caterpillar yield, as compared to the control.

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*This work was done jointly with the All-Union Scientific Research Institute of Fermentation Products of the USSR Food Industry.

**This work was done jointly with the Institute of Developmental Biology imeni N. K. Kol'tsov, USSR Academy of Sciences, under the supervision of V. A. Strupnikov, corresponding member of the USSR Academy of Sciences.

Let us now turn to experiments dealing with the effects of mm radiation on animals [29-37]. Experiments were conducted with low power flux density (9-10 mW/cm^2) to demonstrate the specific effects of this form of radiation. It was of great interest to assess the depth of penetration of SHF radiation in the mm range in experimental animals. Analysis of the obtained distribution of radiation energy in the skin of mice revealed that about 70% is absorbed in the dermal layer, where there is a rather dense network of nerve endings. This warranted the assumption that mm waves may only have a mediated effect on an organism [34]. Experiments also established that mm waves cause no overt disturbances in animals, and are not instrumental in growth of malignant neoplasms [35]. At the same time, when SHF radiation is combined with x-rays or antineoplastic agents (fluorafur, sarcolysin, cyclophosphane, imiphos), electromagnetic waves are capable of lowering significantly the deleterious effect of these factors on bone marrow hemopoiesis [36]. It must be noted that, when combining mm waves with x-radiation, the order of these factors was important. Thus, when x-radiation preceded mm waves there was enhancement of the depressing effect of the first agent. When the order was changed (SHF + x-radiation), there was significant attenuation of the deleterious effect of ionizing radiation on bone marrow cells [29]. A rather high coefficient of protection (K = 2-3) of bone marrow cells was obtained (Figure 2).

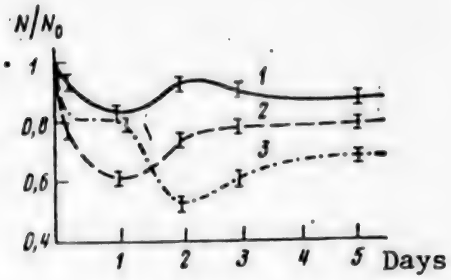

Figure 2.
Biological effect as a function of time of exposure to combined factors. X-axis, time, days; y-axis, ratio of postexposure bone marrow cells to number of such cells in the control.

1) SHF and x-radiation
2) x-radiation
3) x-radiation and SHF

Analysis of the results of SHF combined with antineoplastic agents established that mm waves also attenuate significantly the deleterious effect on bone marrow. For example, there was a 2.5-fold decrease (60% deficiency) in number of bone marrow cells on the 3d day after giving animals a therapeutic dose of the antineoplastic agent, cyclophosphane, whereas in the group of SHF + cyclophosphane the decline constituted 25-30% at this time [31]. The same findings were made with the use of SHF combined with other antineoplastic agents on bone marrow cells [30, 32, 33]. Studies of hemopoiesis in the combined group and under the influence of antineoplastic agents or x-radiation, separately, demonstrated that SHF radiation in the mm range selectively affected different classes of hemopoiesis. This selectivity was related to the combination used. Thus, with some combinations (SHF + chrysomallin or x-radiation) there was significant preservation of myeloid and erythroid cells. There was drastic increase in parent cells and erythroid cells with the combination of SHF and fluorafur. It must be also noted that, with the combination the mitotic index remained normal or somewhat above normal, whereas with the use of the agent or x-radiation alone, it was below normal and recovery occurred by the 5th-10th day after the treatment [32, 33, 36].

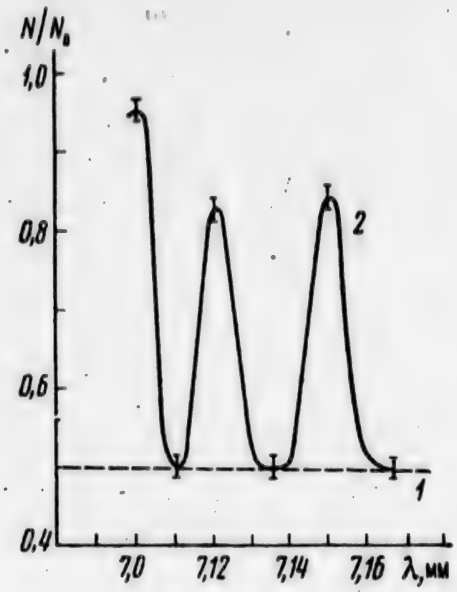

Figure 3.
Biological effect (N/N_0) as a function of wavelength λ; N/N_0--number of bone marrow cells after treatment as related to normal number of cells

1) x-radiation
2) SHF and x-radiation

In conducting experiments it was interesting to determine the biological effect (protection of bone marrow cells) as a function of intensity of SHF radiation, time of exposure and wavelength. A certain threshold value of power flux density was found (~10 mW/cm^2), below which the SHF field had no effect. Further increase in intensity did not alter the obtained biological effect. As for exposure time, variation thereof revealed that 60-min exposure was the optimum. However, it must be mentioned that the biological effect did not increase with increase in irradiation time [37]. It was of the utmost interest to examine the biological effect as a function of SHF wavelength in experiments using the combination of SHF and x-radiation. The wavelength of SHF was changed by 0.01 mm (Figure 3). It was found that the relative number of remaining bone marrow cells increased from 0.50 to 0.85-0.95 at certain values of λ in the range of 7.09-7.16 mm at wavelengths of 7.095, 7.1, 7.12 and 7.15 mm, whereas there was no protective effect at all with other wavelengths (7.11, 7.13, 7.14, 7.16 mm)[35]. Such strict dependence of the biological effect on wavelength is of a resonance nature with bandwidth of $\Delta f/f \sim 2 \cdot 10^{-3}$. A maximum biological effect was observed with λ~7.1 mm, whereas a change of 0.01 mm (0.16%) in wavelength led to total disappearance of the phenomenon.

It is deemed very important to study the effect of mm radiation on tumor growth. To date, data have been obtained that are indicative of significant inhibition of growth of a transferred tumor (rat sarcoma 45) in the case of the combined and successive effect of x-radiation and mm waves. According to the results of these experiments, there is significant (2-2.5-fold) inhibition of tumor growth under the combined effect of x-radiation and mm waves in a specific order, as compared to the effect of x-radiation alone. The mechanism of action of mm waves in these experiments was even more complex than in the experiments with microorganisms. The fact of the matter is that the depth of penetration of radiation into tissue with this frequency range is about 300 μm, i.e., one could hardly refer to the direct effect of mm radiation on bone marrow cells. Moreover, it was established that only when specific parts of the animal's body are exposed to mm waves do they manifest a protective effect which, however, extends over the entire body. These facts indicate that we are dealing here with some sort of abscopal [mediated] interaction between mm radiation and the hemopoietic system.

Conclusion

Thus, specific nonthermal electromagnetic waves in the millimeter range may have a strong and selective effect on the nature of development and vital functions of organisms and biological structures differing in complexity. Use of the results of exposure to this factor may have important practical implications. But, although quite a bit has been accomplished to date, we are still at the start of our journey. First of all, there is no strict or complete theory concerning the observed phenomena. Nor has there been practical introduction as yet, although there are several obvious practical applications. For this reason, it is deemed purposeful to intensify work in this new and promising direction. It must be noted that the original research conducted under the guidance of S. Ye. Manoylov, V. F. Kondrat'yeva, Ye. N. Chistyakova, R. I. Kiselev, N. P. Zalyubovskaya and others has not been reflected in this article.

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CURRENT PROBLEMS OF RADIOBIOLOGY OF ELECTROMAGNETIC RADIATION OF RADIO-FREQUENCY RANGE

Moscow RADIOBIOLOGIYA in Russian Vol 20, No 1, Jan-Feb 80 pp 3-8

[Article by I.G. Akoyev, Institute of Biological Physics, USSR Academy of Sciences, Pushchino]

[Text] On the basis of analysis of the current state of radiobiology of nonionizing radiations, the need of accelerated development of basic research of the biological action of these radiations, especially in the radio-frequency range is substantiated. An objective is set for accumulation of experimental data on questions of reversibility and cumulation of effects, selectivity, specificity and additivity, direct and indirect action, sensitivity, adaptation and stability in regard to radiation, species differences and possible mechanisms of operation of radiation.

Up to the present time, radiation biology has concentrated its chief attention on the investigation of the biological effect of radiations of that part of the electromagnetic spectrum which is ionizing and encompasses frequencies from 10^{17} to 10^{27}. This is a band of frequencies stretching from ultraviolet and soft x-ray radiation to the ultrahard components of cosmic radiation.

But recently in connection with the achievements of the scientific-technical revolution, an acute need has arisen of knowing the biological action of nonionizing electromagnetic radiations (frequencies of 10^{0}-10^{16} Hz), especially in the radio-frequency range (1,2). The introduction to science, medicine and the national economy of technical aids and processes, utilizing electromagnetic fields, is bringing about an exponential growth of irradiation of people. Significant contingents of the population of the United States are being subjected constantly at the present time to irradiation with electromagnetic fields in the radio-frequency range in doses exceeding the permissible levels adopted in the USSR. Further technical progress presupposes the use of new technical aids connected with the use of powerful magnetic and electromagnetic fields (thermonuclear and space power stations, telecommunications, radar, communication and navigation equipment, superhigh voltage electric transmission lines, household superhigh-frequency ovens and so on), which will expand the scale and intensity of action of nonionizing electromagnetic radiations on man and the biosphere as a whole (1, 2). At the same time, it has been shown that the action of these radiations posses a biological effect (3-6). Depending on the radiation dose, there may be observed transitory functional changes, morbid states and even unfavorable outcomes. They are also being used for therapeutic purposes (ultra-high-frequency therapy).

Despite a large number of published works, the level of knowledge of the biological action of nonionizing electromagnetic radiations (NI EMR) is inadequate. Basic studies with rare exceptions are conducted disconnectedly, in the majority of cases by isolated small groups and frequently on the basis of conflicting current requirements at the methodological level. The mechanisms of operation have not been studied. Representations of safe levels vary in different countries on the order of 2-3 (2, 3). The level of knowledge of the biological action of nonionizing radiations is not in accord with the demands of the present technical revolution and is becoming a hindrance to the introduction to the national economy of the latest technical aids and processes connected with the use of nonionizing electromagnetic radiations and magnetic fields. Accelerated development of research on the biological action of nonionizing radiation is a social command upon a modern society.

But it would be extremely difficult to solve these questions because of the tremendous range of frequencies (of more than 15 orders), essential differences in the relation of leading physical processes in different frequency intervals, endless diversity of alternatives of amplitude and frequency modulation, lack of knowledge of species peculiarities of effect on animals and ways of extrapolation of experimental results in regard to the human organism (2, 5, 7). This requires a basic investigation of the general laws of operation of these radiations, a knowledge of the leading mechanisms of interaction of the absorbed energy with biological structures and serious theoretical research.

Certain Features of Biological Action

The biological action of electromagnetic radiations of the radio-frequency range is examined in detail in a number of surveys (2-7). The difficulties of generalizing existing data consist of an inadequate description of the physical conditions of the experiment or clinical case. The data are frequently contradictory and difficult to compare, especially in the case of those obtained in the earlier years. In the last technical report-survey on biological effects of nonionizing radiation presented in May 1978 by a group of leading U.S. specialists in the service of policy planning in science and technology for the U.S. president (2), it is said outright that in view of the complexity of the problem and the limitedness of information on possible versions of physical conditions of operation, it would be impossible to make a definitive biological conclusion. It is enough to point out, for example such methodological difficulties as the unsuitability of generally accepted electrophysiological methods with metal electrodes and conductors, significantly changing the field of radiation, general and local doses of absorbed energy (5, 7). For this reason the methods of securing animals and radiation chambers with metallic parts were unsuitable. A change of the bodily position of an animal as to angle, perpendicular direction of emission could change the total dose of absorbed energy by a factor of 1-2. These are the complaxities of the methodological plan, hindering the conducting of biological experiments with reproduceable conditions of irradiation.

The accumulated data, however, make it possible to speak of the qualitative features of the biological effects caused by electromagnetic radiations of radio-frequency range. Let us examine them from the point of view of a solution of general biological problems determining the basic specific nature of the given type of pathology.

Reversibility of occurring changes. The chief feature of the effect of radiations is the reversibility of the absolute majority of changes occurring in an irradiated organism. This has been traced by clinicians. It is borne out by experiments on animals and model systems. Reversibility is light and quick. Morphological changes are either lacking or weakly expressed. It is enough for an irradiated person to have a brief leave for most of his complaints to disappear (5). In experiments on artificial phospholipid membranes modified by channel-forming antibiotics, changes in ionic conductivity occur from the time of turning on the radiation generator and return to normal within a minute of it being turned off (8, 9). But irreversible changes are also known. They were manifested in the inception of foci of cataract genesis, in irreversible chromosome disturbances of cells, in genetic changes of microorganisms (3, 5-7).

Possibility of adaptation. Observations of the reaction of the organism and its systems to repeated irradiations disclosed in a number of cases the possibility of adaptation to nonionizing radiation. Each subsequent irradiation, separated from the preceding action by an insignificant interval of time, was borne easier than the first irradiation. It is interesting that even a rise in bodily temperature in response to the action of the radiation in large doses was significantly smaller with repeated irradiations (10). But the limits of development of adaptive states of the organism and the basic conditions and factors making easier or more difficult adaptative reactions are unclear.

Possibility of cumulation of effects. With chronic action in the form of such small daily doses, each of which clearly does not cause any sort of biological reactions, biological reactions begin to appear on the expiration of a certain period of irradiations as the result of cumulation of unobserved previously latent changes. The clinical observations on this level are most characteristic (5). Experimental data are also to be found. Thus, in experiments on guinea pigs involving daily 7-hour irradiation with a very low density of current with a power of 1-50 microwatts per square centimeter, it was noted that during the course of the first three weeks of irradiations no changes of any kind occurred, including from the direction of the absolute phagocytic index of blood neutrophils. But beginning with the fourth week, this indicator began to change in a statistically significant manner (11).

Selectivity of action. It is not clear whether there are organs and tissues that are especially sensitive to nonionizing electromagnetic radiations with equal doses of absorbed energy. The question of the selectivity of action of nonionizing electromagnetic radiations has not been studied at all. It is noted that with great intensity of irradiation of the organism the biggest pathological changes occur in the crystalline lens of the eye, the testes and the mucosa of the stomach and the intestinal tract (4, 5). The applicability of the law of Bergonier and Tribondo has not been elucidated. The greater affection of these organs may be connected not only with the special features of proliferation but also with a special feature of blood supply (removal of heat) as well as with differences in the dielectric constant and conductivity of the tissues comprising them. The question of selectivity of action is closely connected with the problem of critical organs for a given type of action without the knowledge of which it is impossible to determine maximum permissible levels of action.

Additivity with other actions. The additivity of action of nonionizing electromagnetic radiations of the centimeter range and of infrared rays was experimentally shown, but is partial.

Even in the case of an identical elevation of bodily temperature by these actions, differences exist both in external manifestations and also in the character of subsequent adaptive reactions (4, 5). Special interest is presented by the combination of the actions of nonionizing and ionizing electromagnetic radiation. Depending on the order of application of these physical factors, there are produced both a reciprocally aggravating action and a protective action (4-7). The investigation of these questions is only getting under way, and the necessary data have as yet not been obtained. They are especially important for the therapy of neoplastic diseases.

Direct and mediated action. With the application of penetrating nonionizing electromagnetic radiations on the organism, it is customarily believed that such effects as cataracts, stomach ulcers and atrophies of the testes are the result of the direct action of absorption of energy by these organs and are connected with the process of heat emission. For other effects such as neuroses, asthenia, cardiovascular functional changes, especially in the case of chronic nonheat effects, it is more difficult to determine with what form of action they are connected. For nonpenetrating electromagnetic radiations of the millimeter range (depth of penetration is less than 1 mm), general effects are disclosed (for example, changes in the number of hemopoietic cells of the bone marrow), which are manifestly developed through neuroendocrine regulation (12). Recently new data were obtained on the higher sensitivity of such effects mediated through neuroendocrine regulation compared to the effects of indirect action. Radiation doses for eliciting chromosome aberrations of cells of culture of lymphocytes were significantly smaller in the case of operation of nonionizing electromagnetic radiations on the whole organism than on the culture of these same cells in vitro (3).

Specific action. Even in early works, the supposition was stated that for penetrating nonionizing electromagnetic radiations there is in addition to a basic heat action a specific action (4). As proof of the existence of specific action, data are frequently cited concerning the absence of elevated bodily temperature or incomplete additivity with heat action caused by other physical factors. This question has been weakly developed. First of all, there should be kept in mind, the special features of distribution of the heat generated in the organism. It could be most unequal. Furthermore, a 5°C change in the temperature of an organ changes rectal temperature by less than 0.1°C. This irregularity in distribution of heat is also characteristic of biological ultrastructures. For example, in model experiments, the temperature of a biological membrane can be several orders less than the temperature of the ionic channel in it. Consequently, it is impossible to judge the specifics of action in the absence of changes in the measured temperature of the body or organ. At the same time, experimental materials exist on the occurrence of biological reactions in the action of radiation of such extremely low density of power flux (1-10 micro watts per square centimeter) that it becomes evidently difficult to assume a significant role for the release of heat (4, 5).

Species differences. Knowledge of them is important for the solution of questions of extrapolation of experimental data from animals for man, but they have been completely unstudied. If for penetrating radio-frequency radiations heat release be considered as the basis of their biological action (this is undoubtedly true of large doses), then the capacity of the organism to withstand internal local and general overheating must have a certain dependency, in our opinion, on the intensity of heat and energy metabolism inherent in the given species of warm-blooded animals. Available information on the high level of affection of small mammals is connected with lack of consideration of the average dose of absorbed energy per gram of tissue in such experiments. Tissue, for example in radiation with 10-cm waves will vary significantly in mice and dogs, since this radiation is of low penetration for dogs (depth of penetration is roughly equal to one-tenth of the radiation wave length).

Resistance and sensitivity to radiation. In the radiobiology of nonionizing electromagnetic radiations, the principles and criteria of evaluating resistance to radiation have not been worked out. Most researchers identify sensitivity and resistance to radiation. The appearance of any biological reactions to radiation is still frequently considered as an indicator of exceeding of the permissible radiation dose. In this evidently is to be found one of the causes of the existing significant differences in permissible levels of action in different countries, since use is made of different methodological approaches.

There have as yet not been accumulated data that would make it possible to separate out from the totality of manifestations of biological action of electromagnetic radiations signs of development of nonspecific adaptive reactions of varying strength, and they evidently can be of major importance, especially in weak effects. There have as yet not been obtained quantitative relations of expression of biological effects on density of power flux and other physical conditions of operation. These questions have turned out to be very complex. It is sufficient to say that under certain conditions, "windows" were revealed of higher biological effectiveness in the case of power flux density that is specific and intermediate in size (13).

On Possible Mechanisms of Biological Action

It would be possible to speak more specifically of the mechanisms of biological action of nonionizing electromagnetic radiations only after study of the physical nature of the initial processes of the interaction of radiations with biomolecules and study of subsequent processes of transformation of occurring changes down to physiological reactions. But up to the present time, these processes have been little studied.

Heat release is the leading physical process in the action of penetrating radiation emissions in medium and large doses. For nonpenetrating radiation emissions, the possibility was shown of resonancelike intermittent pulsing effects. In the operation of low-frequency and static fields, a leading role may be played by magnetic and electric fields. A number of suppositions has been made on primary physical processes during the operation of nonionizing electromagnetic radiations, not one of which has been strictly proved. In primary reactions, intensification of oscillations of molecules and ions is supposed, as well as an increase in their kinetic energy and mobility, their definite orientation and participation of bound water and free-radical processes (3, 7). The possibility is emphasized of cooperative conformational rearrangements, changes in permeability and other properties of biological membranes, disturbances connected with calcium of processes on membranes (13, 14). There was experimentally shown the influence of decimetric waves on the operation of ion channels produced on artificial phospholipid membranes and on the model postsynaptic membranes (8, 9). Nonionizing electromagnetic radiations can affect the rhythm of cellular and physiological processes; at the same time low-frequency "windows" were revealed for such indicator as deviations in EEG or flow-off of calcium from brain tissue (13).

The opinion has been voiced that maximum sensitivity to nonionizing electromagnetic radiations is possessed by whole organisms, less sensitivity by isolated organs and still less by molecules (4, 5). For this reason, for example, a cellular system is more sensitive to the action of radiations in vivo than in vitro. This is connected with the significant role of reactions mediated through the neurohumoral system, whose sensitivity it is opined (14) is due to the special structure of the conducting pathways of the hippocampus of the brain, which focus heightened absorption of energy in this structure.

Basic Objectives

In the radiobiology of nonionizing electromagnetic radiations, the period of accumulation of phenomenological data is not yet over. Obtaining them is necessary for the study of the special features of the biological action of this form of radiation. Special attention should be directed to raising the scientific-methodological level of conducted research, especially from the point of view of the correctness of the physical conditions of conducting the experiment. Generalization of accumulated data, their theoretical analysis for the purpose of determining the general laws of operation of nonionizing radiations should occupy a correspondingly important place. The most important part of the problem at the present time is the investigation of possible mechanisms of action of nonionizing electromagnetic radiation on biological subjects, proof of existing assumptions concerning them and proposal of new ones.

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PARTICULAR FEATURES OF CONDITIONED ELECTRODEFENSIVE REFLEX IN WHITE RATS ON BACKGROUND OF CONSTANT MAGNETIC FIELD

Kiev FIZIOLOGICHESKIY ZHURNAL in Russian Vol 26, No 2, Mar-Apr 80 pp 264-268

[Article by I. V. Shust, S. I. Galantyuk and Yu. V. Cheretyanko]

[Text] Study of the influence of magnetic fields upon the higher nervous activity of man and animals has long been attracting the attention of researchers. It is indicated in the literature that magnetic fields inhibit development of conditioned reflexes in planarians (10), fishes (2,17), and mammals (3,16,17). However, there are data of opposite nature as well (5,18), indicating accelerated development of the avoidance reflex in animals exposed previously to a magnetic field.

We studied formation of a conditioned electrodefensive reflex (CER) in white rats exposed to a constant magnetic field (CMF), and the influence of a vitamin preparation--galascorbin--on formation of the CER in animals exposed to a CMF.

Research Methods

The experiments were conducted on male rats weighing 160-190 gm using a motor electrodefensive technique (9) modified by the authors. Conditioned reflexes were developed in a specially built chamber, on calm animals; silence and other necessary conditions were observed. The chamber consisted of two parts separated by a partition; the latter had a round hole with a diameter of 6.5 cm located 15 cm above the floor, which was made from electrode wire. An electric 40 watt lamp was used as the conditioned stimulus, and a weak alternating current, the threshold intensity of which was set by a laboratory autotransformer, was used as the unconditioned stimulus. The threshold of the animals' avoidance reaction was measured with a TL-4 instrument. Following electrocutaneous stimulation the rat jumped through the hole in the partition into the other part of the chamber, where it was once again exposed to the stimuli. Each of the rats was subjected to 30 combinations daily with an interval of 20 sec; the unconditioned stimulus was turned on at the 6th second of action of the indifferent stimulus. The time of the joint action of the conditioned and unconditioned stimuli was 10 sec. The tenacity of the developed CER was calculated as a percentage ratio between the number of positive motor reactions to the conditioned stimulus and the total number of combinations presented.

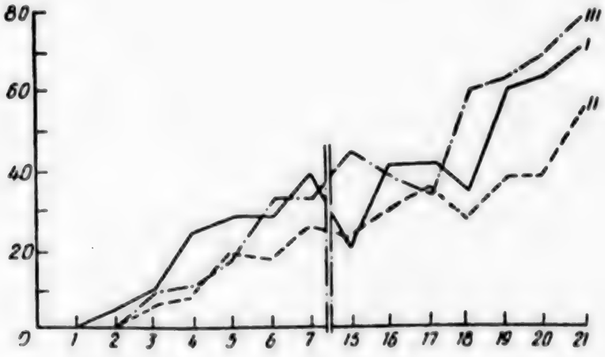
Animals used in the experiment were divided into three groups of four rats each. Group I served as a control. Rats in groups II and III were exposed to a uniform 1,000 Oe constant magnetic field in daily sessions of 6 hours for 1 week. Animals in group III were given 10 percent aqueous galascorbin solution perorally for 7 days, at a dosage of 100 mg of the preparation per 100 gm live weight; rats in groups I and II received the corresponding quantities of distilled water. During the time that animals in groups II and III were within the CMF, the control group was kept in a zone in which the magnetic field intensity did not exceed 1 Oe.

Animals in each group were exposed to the factors indicated above and allowed to develop the CER for 7 days, after which, following a 1-week interval the CER was developed in the rats for a period of 7 days without subjecting them to the influence of the CMF and galascorbin in order to study the aftereffects of the CMF.

The numerical data obtained were treated by the method of variational statistics, as described by I. A. Oyvin (1960).

Research Results and Discussion

The experiments showed that exposure to a 1,000 Oe CMF for 6 hours daily for 1 week reduces the tenacity of the CER in rats throughout practically the entire experimental time. It is evident from Table 1 that animals in group II were unable to develop the CER on the second day, some of them (No 7 and 8) were unable to do so on the third day, while rat No 7 was not even able to do so on the fourth day of the experiments, while in the control group the tenacity of the CER on the second and third days was 5.0 ± 1.87 and 10.2 ± 5.00 percent respectively.



Formation of a conditioned electrodefensive reflex in control rats (*I*), rats exposed to a CMF (*II*), and rats exposed to galascorbin and a CMF (*III*): The tenacity of the CER is shown on the vertical axis (percent), and experimental days are shown on the horizontal axis.

In addition, beginning with the first days of the experiment, growth in the threshold of the avoidance reaction was noted among rats in group II, which were first subjected to a CMF (Table 2); this is an indication of reduced excitability of the skin receptors of the animals. This result is consistent with the published data (21) indicating change in autonomic reactivity of the body in response to one-time exposure to a weak CMF for 1 hour. By the end of the experiment the threshold indicators for the avoidance reaction of the rats of this group attained the level typical of the control group, which can obviously be interpreted as a manifestation of adaptation to CMF exposure. A similar conclusion was reached in research on the influence of a nonuniform 150-200 Oe CMF on formation of temporary associations in the rat cerebral cortex (6).

Table 1. Effect of a Constant Magnetic Field and Galascorbin on Development of a Conditioned Electrodefensive Reflex (Percent) in White Rats

Группа крыс (1)	Условия опыта (2)	Номер крысы (3)	(4) Дни опыта													
			1	2	3	4	5	6	7	15	16	17	18	19	20	21
(5) I	Контрольные крысы	1	0	10,0	26,7	40,0	46,7	46,7	73,3	26,7	36,7	50,0	60,0	73,3	73,3	76,7
		2	0	3,3	6,7	30,0	23,3	23,3	26,7	20,0	46,7	30,0	10,0	16,7	66,7	86,7
		3	0	0	0	13,3	13,3	16,7	16,7	3,3	46,7	33,3	33,3	60,0	46,7	60,0
		4	0	6,7	6,7	13,3	30,0	30,0	36,7	33,3	33,3	53,3	33,3	86,7	70,0	56,7
		M	0	5,0	10,2	24,2	28,3	29,2	38,4	20,8	40,8	41,6	34,2	59,2	64,2	70,0
		±*m*		1,87	5,00	5,70	6,08	5,57	10,69	5,57	2,98	5,06	8,85	13,14	5,18	6,12
(6) II	Крысы, которых подвергали воздействию ПМП	5	0	0	3,3	0	10,0	10,0	6,7	13,3	3,3	6,7	6,7	10,0	3,3	0
		6	0	0	23,3	30,0	46,7	53,3	76,7	53,3	66,7	96,7	86,7	83,3	86,7	96,7
		7	0	0	0	0	6,7	0	10,0	13,3	43,3	26,7	6,7	26,7	26,7	46,7
		8	0	0	0	3,3	10,0	3,3	10,0	10,0	6,7	10,0	6,7	16,7	33,3	66,7
		M	0	0	6,6	8,3	18,4	16,7	25,8	22,5	30,0	35,0	26,7	36,7	37,5	52,5
		±*m*			4,85	6,29	8,21	10,73	14,69	8,92	13,18	18,2	17,32	14,5	15,2	17,58
		t		2,67	0,37	1,87	0,97	1,03	0,69	0,16	0,80	0,64	0,39	1,15	0,71	0,18
(7) III	Крысы, которым вводили галаскорбин и подвергали воздействию ПМП	9	0	0	6,7	6,7	20,0	30,0	20,0	26,7	33,3	33,3	50,0	46,7	56,7	80,0
		10	0	0	3,3	10,0	13,3	26,7	33,3	33,3	40,0	36,7	46,7	60,0	70,0	76,7
		11	0	0	23,3	23,3	26,7	66,7	53,3	50,0	50,0	66,7	73,3	100,0	66,7	73,3
		12	0	0	3,3	6,7	10,0	6,7	23,3	26,7	30,0	16,7	66,7	43,3	80,0	83,3
		M	0	0	9,2	11,7	17,5	32,5	32,5	44,2	38,3	38,4	59,2	62,5	68,4	78,3
		±*m*			4,14	3,42	3,21	10,82	6,49	6,91	3,82	9,01	5,57	11,26	4,16	1,86
		t		2,67	0,41	0,48	0,10	1,04	0,42	1,92	0,60	0,18	1,79	1,40	1,96	1,46

Key:

1. Rat group
2. Experimental conditions
3. Rat number
4. Experimental day
5. Control rats
6. Rats exposed to CMF
7. Rats given galascorbin and exposed to CMF

Table 2. Change in the Stimulation Threshold of Rats in Response to a Constant Magnetic Field

(1) Дни опытов	Пороговое напряжение тока (*B*)* (2)		*p*	Дни опытов (1)	Пороговое напряжение (2) тока (*B*)		*p*
	Контроль *M*±*m*	Опыт *M*±*m*			Контроль *M*±*m* (3)	Опыт *M*±*m* (4)	
1	32±1	40±1	<0,001	15	35±1	25±3	<0,02
2	30±1	36±0	<0,001	16	30±2	26±2	0,2
3	30±1	37±1	<0,001	17	30±1	27±2	<0,2
4	30±1	30±2	—	18	30±0	26±3	<0,5
5	30±1	30±1	—	19	30±1	30±2	—
6	29±2	34±2	<0,2	20	30±1	30±2	—
7	36±1	30±1	<0,01	21	30±1	30±2	—

Key:

1. Experimental days
2. Current threshold voltage (v)*
3. Control
4. Experiment

*The threshold current voltage was measured and, correspondingly, the data were subjected to mathematical treatment with an accuracy of ± 1 v.

In the period of the aftereffect of the CMF, beginning with the 16th through 21st days of the experiment, the tenacity of the CER remained low in comparison with the control (see figure); however, there were no statistically significant differences.

As we know, formation of a temporary association lies at the basis of conditioned reflex formation. According to Roytbak's hypothesis (13), establishment of the association is the result of morphological alterations in the nervous system. A major role in this is given to neuroglial cells, which have a direct part in myelinization of nerve fibers. It appears highly probable that change in the conditioned reflex activity of the animals in response to the CMF is associated with morphological disturbance of the neuroglia which, as was demonstrated by some researchers (1,7,14,20), is highly sensitive to magnetic fields. It should be noted that neurons may retain their morphological integrity when neuroglia reacts to a CMF (4).

Morphological changes in the central nervous system persist for a long period of time following cessation of the action of a magnetic field (17); this is consistent with our experiments, in which the tenacity of the CER developed in rats of group II was found to be low in the period of the aftereffect of the CMF--that is, from the 15th through 21st days of the experiment.

Another factor that may contribute significantly to disturbance of formation of a CER to a light stimulus is the decrease is excitability of the cortical division of the visual analyzer, noted in experiments on rabbits exposed to a 200 Oe CMF (17); metabolic changes occurring in eye tissue in response to a magnetic field may also be important (11).

We attempted to use a vitamin preparation--galascorbin--to reduce the negative influence of the CMF on formation of the conditioned reflex. This preparation improves carbohydrate-phosphorus metabolism (12), diminishes the injurious action of radiation (15) and, as we had revealed earlier (8,19), it has a normalizing influence upon the liver of white rats exposed to an intense CMF.

The obtained data (see figure and Table 1) indicate that galascorbin raises the tenacity of the CER. Thus during almost all days of the first experimental week the tenacity of the CER was greater among animals of group III than among animals of group II, which were also exposed to a CMF but were not given galascorbin. We observed the preparation to have pronounced action during the CMF aftereffect period, from the 18th through 21st days of the experiments. In this case the tenacity of the CER developed by rats in group III increased even in comparison with the control.

Judging from the biological role played by galascorbin in metabolism, we may hypothesize that the normalizing influences of the preparation upon development of the CER in rats during their time of exposure to a CMF and in the period of its aftereffect is associated with intensification of oxidation-reduction processes in neuroglial cells and in neurons, and consequently with improvement of consolidation of short-term memory.

Conclusion

1. Exposure to a 1,000 Oe CMF reduces the tenacity of the conditioned electrodefensive reflex in white rats, and to a greater degree in the initial phase of its development.

2. Changes in conditioned reflex activity are observed not only during the time of exposure to a CMF but also in the aftereffect period.

3. In the presence of a CMF with the indicated parameters, the threshold of the avoidance reaction of the rats initially increases, and then it decreases, after which normalization of this indicator is observed until the end of the experiment.

4. Administration of galascorbin to the animals promotes tenacious development of the conditioned reflex, which remains more-pronounced in the CMF aftereffect period.

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MORTALITY IN ACUTE MYOCARDIAL INFARCTION AS FUNCTION OF GEOMAGNETIC FIELD ACTIVITY IN YEREVAN

Yerevan DOKLADY AKADEMII NAUK ARMYANSKOY SSR in Russian Vol 22, No 3, 1981
pp 188-191

[Article by N. N. Karazyan and L. A. Akhverdyan, Yerevan State Medical Institute, Institute of Geophysics and Engineering Seismology, Armenian SSR Academy of Sciences]

[Text] At a time of intensive mastery of the cosmos, there is no longer any doubt that many natural phenomena on Earth are a function of cosmic conditions (1). Study of the character of geomagnetic influences on living organisms is one of the new and promising directions of biology (2).

Measurements taken on the Elektron-4, Explorer-26, Alouette-2, Kosmos-26 satellites gave rise to completely new ideas about the sources of magnetic storms and made it possible to use observatory data in other fields of science. The time came for the arisal of the issue of scientific collaboration between geophysics and medicine in order to establish a reliable correlation between biomedical and geophysical phenomena.

Clinical experience has demonstrated that mortality from myocardial infarction undergoes significant fluctuation on different days. In recent years these fluctuations have been linked to solar activity and disturbances in the geomagnetic field.

In their studies most authors discovered a direct association between the activity of the magnetic field of Earth and cardiovascular accidents, in particular, mortality from myocardial infarction (3-9). At the same time, the magnitude and form of geomagnetic variations are considered to be functions of the latitude of the place of observation, the former increasing from the equator to the pole (10). Other authors, however, deny such an association (11).

Such research has not been conducted in the Armenian SSR, consequently we set ourselves the task of studying the influence of geomagnetic field activity on mortality from myocardial infarction in the city of Yerevan. We used the data of the average daily fluctuation of the K-index to evaluate the geomagnetic field. Days when $K \leqq 2.5$ are considered magnetically quiescent; days when $K=3$-3.5, days of average activity, and days when $K \geqq 4$, magnetically active.

We analyzed the medical history of patients who died from acute myocardial infarction in therapeutic and specialized cardiological divisions of a number of clinics in the city of Yerevan during 1974-1978 with the objective of establishing the correct date of death, age and sex of the deceased, as well as the features of

Table 1. Mortality from Acute Myocardial Infarction As a Function of Geomagnetic Field Activity

(1) Геомагнитная характеристика дней	(2) Число дней	(3) Число смертных случаев	(4) Ежесуточная смертность (M±m)	(5) Степень достоверной оценки	
				t	*P*
(6) Магнитоспокойные дни(K $<2{,}5$)	1303	343	0,26±0,015	—	—
(7) Дни умеренной активности(K $= 3{,}35$)	154	52	0,34±0,05	1,5	<0,25
(8) Магнитоактивные дни(K >4)	257	123	0,48±0,046	4,6	<0,001
(9) Постактивные дни	112	53	0,47±0,075	2,8	<0,02

(10) *Примечание.* P вычислено по сравнению с показателями магнитоспокойных дней.

Key:

1. Geomagnetic description of day.
2. Number of days
3. Number of deaths
4. Daily fatality (M±m)
5. Degree of reliability of evaluation
6. Magnetically quiescent days ($K\leq 2.5$)
7. Days of moderate activity ($K=3.35$)
8. Magnetically active days ($K\leq 4$)
9. Postactive days
10. Note. Rho is calculated from the indices for magnetically quiescent days.

the course of the disease. The causes of death in each individual case were determined on the basis of clinical and pathoanatomical data. Also considered were cases of infarction with fatal outcomes ("home fatalities") from the call sheet of the central first aid station for the same period in the city of Yerevan. In all cases the diagnosis of myocardial infarction was determined by the physicians of anti-infarction teams and confirmed electrocardiographically.

During the same period the Garni observatory recorded 88 days with brief (less than 24 hours) magnetic disturbances and 56, with prolonged (2 days or more) magnetic disturbances: 169 days in all.

As our research demonstrated, brief magnetic disturbances during the two previous and three subsequent days did not influence the mortality rate from myocardial infarction. At the same time, prolonged magnetic disturbances, which did not influence the indices of the two previous and third subsequent days, produced a significant increase in the mortality rate during the two subsequent days, which were placed into a separate group (112 days) and conventionally designated "postactive days".

Of 571 deaths investigated by us, 425 (74.4 percent) were men and 146 (25.6 percent) women. At the same time, for ages 0 to 49 years (group I) there were 101 cases; 50 to 59 years (group II), 109 (19.1 percent); 60 to 69 years (group III), 226 (39.6 percent); 70 years and older, (group IV), 135 (23.6 percent).

As table 1 shows, the daily mortality rate on magnetically active, postactive and moderately active days is higher than on magnetically quiescent days.

The ratio between the indices of average daily mortality on magnetically quiescent days and the indices on active, postactive and moderately active days were 1:1.85, 1:1.81 and 1:1.31, respectively, the indices on active and postactive days being reliable (according to Student's t).

A similar pattern was detected during annual analysis of mortality.

Our studies demonstrated that while on magnetically quiescent days the average daily mortality in groups I, II, III and IV was (in percent) 0.044±0.006; 0.055± 0.006; 0.107±0.009 and 0.058±0.007, on magnetically active days it was equal to 0.101±0.02; 0.082±0.02; 0.175±0.028 and 0.121±0.25 cases, respectively. The ratio of the indices of average daily mortality on magnetically quiescent and magnetically active days was 1:2.3 for group I, 1:1.49 for group II; 1:1.04 for group III and 1:2.09 for group IV, i.e., patients younger than 50 and older than 70 years displayed great sensitivity to geomagnetic activity. An analogous picture was revealed on postactive days. On moderately active days the greatest sensitivity was displayed by patients older than 70.

Our observations further indicated that male patients proved to be more sensitive to the activity of the magnetic field of Earth than female patients. Thus, while on magnetically quiescent days the average daily mortality in men was 0.198±0.013, and in women 0.065±0.007, on magnetically active days it was 0.374±0.042 and 0.105± 0.021, respectively. The ratio of the indices of average daily mortality on magnetically quiescent and magnetically active days is 1:1.89 in men, and 1:162 in

Table 2. Lethal Complications As Function of Geomagnetic Field Activity

Зависимость летальных осложнений от активности геомагнитного поля

(1) Характер летального осложнения	(7) Всего		(8) В спокойные дни		(9) В умеренно-активные дни		(10) В активные дни		(11) В постактивные дни	
	n = 428		n = 269		n = 33		n = 87		n = 39	
	абс.	%	абс.	% ко всей группе	абс.	% ко всей группе	абс.	% ко всей группе	абс.	% ко всей группе
(2) Сердечная недостаточность	195	45,6	129	47,9	16	48,5	34	39,1	16	41,0
(3) Кардиогенный шок	102	23,8	61	22,7	7	21,2	24	27,6	10	25,6
(4) Фибрилляция желудочков	53	12,4	29	10,8	5	15,2	13	14,9	6	15,4
(5) Разрыв миокарда	52	12,1	35	13,0	4	12,0	9	10,3	4	10,3
(6) Тромбоэмболия	26	6,1	15	5,6	1	3,0	7	8,1	3	7,7

Key:

1. Nature of lethal complication
2. Cardiac insufficiency
3. Cardiogenic shock
4. Ventricular fibrillation
5. Rupture of the myocardium
6. Thromboemolism
7. Total
   n=428
   absolute   percent
8. On quiescent days
   n=269
   absolute   percentage of entire group
9. On moderately active days
   n=33
   absolute   percentage of entire group
10. On active days
    n=87
    absolute   percentage of entire group
11. On postactive days
    n=39
    absolute   percentage of entire group

women.

Analysis of the basic causes of mortality from complications as a function of the condition of the magnetic field of Earth was carried out on data on the hospital mortality rate. It is well known that the basic causes of patient deaths from acute myocardial infarction are coronary insufficiency, cardiogenic shock, ventricular fibrillation, rupture of the myocardium and thromboembolic complications (12, 13).

Analysis of deaths from myocardial infarction revealed that regardless of the activity of the magnetic field of Earth, 45.6 percent of the patients died of coronary insufficiency, 23.8 percent from cardiogenic shock, 12.4 percent, from ventricular fibrillation, 12.1 percent from rupture of the myocardium and 61.1 percent from thromboembolism. But the objective of the present study is to detect the influence of the activity of the magnetic field of Earth on the frequency of given complications of myocardial infarction resulting in a fatal outcome.

As table 2 shows, on magnetically active days mortality from cardiogenic shock, ventricular fibrillation and thromboembolism is significantly higher than on magnetically quiescent days. Thus, while on magnetically quiescent days 22.7 percent of all those who died from myocardial infarction died of cardiogenic shock, 10.8 percent from ventricular fibrillation and 5.6 percent from thromboembolism, on magnetically active days, deaths from these causes were 27.6, 14.8 and 8.1 percent, respectively. It is interesting to note that mortality from these three types of complications also predominates on postactive days, but on moderately active days mortality from ventricular fibrillation is the highest.

Table 2 gives the impression that on magnetically active and postactive days mortality from cardiac insufficency and cardiorrhexis on a percentage basis is lower than on magnetically quiescent days. It must be noted, however, that it is a question of a relative drop in mortality from cardiac insufficency and rupture of the myocardium owing to an increase in the mortality rate from cardiogenic shock, ventricular fibrillation and thromboembolism on magnetically active and postactive days.

In conclusion, the authors wish to express their deep gratitude to G. O. Badalyan for suggesting the topic and constant help in performing the present study.

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EFFECT OF HYPOGEOMAGNETIC FIELD ON ACTIVITY OF SOME CEREBRAL ENZYMES

Moscow GIGIYENA I SANITARIYA in Russian No 9, Sep 81 (manuscript received 18 Dec 80) pp 11-13

[Article by A. V. Shakula and G. M. Chernyakov]

[Text] At the present time, there is information in the literature concerning the ecological significance of electromagnetic fields of the biosphere, particularly the geomagnetic field (GMF), as environmental factors in the presence of which organisms underwent evolution for many centuries on our planet. Yet, there are some special installations and self-contained units, in which people work for long periods of time, which have shielding properties against the GMF, lowering it by tens of times. For this reason, it is of great interest to hygienists to study the biological effects of a hypogeomagnetic field.

We did not encounter any works in the literature dealing with enzymatic systems of mammals developing under hypogeomagnetic conditions. Our objective here was to examine the activity of the key enzymes of the brain of rabbits whose embryonic and early postnatal development occurred during exposure to a hypogeomagnetic field.

A multilayer cylindrical screen consisting of four layers ["shells"] was used to generate a hypogeomagnetic field. The three inside layers were made of permalloy and were designed to shield animals against stationary and low-frequency (up to several Hz) variable magnetic fields. The fourth (external) layer is made of sheet copper and serves to shield biological objects from electromagnetic fields of industrial origin (50 Hz or more). A ferromagnetic nanoteslameter was used to measure magnetic field induction. The GMF in the experimental chamber was attenuated by an average of 600 times.

Female chinchilla rabbits were taken from the same litter and mated with the same male. Experimental females were placed in the shielded chamber and observed during the periods of gestation (about 30 days), birth and development of baby rabbits to the age of 1 month. During the same period of time, control females were kept in a polyvinyl chloride plastic chamber where the magnetic conditions corresponded to GMF induction at the latitude of Leningrad (about 50 μkT). Air temperature and humidity, diet and other hygienic conditions were identical in the experimental and control groups of animals. The rabbits (16 experimental and 16 control) were decapitated 1 month after birth. Pieces of brain, 1×1×1 cm in size from the frontal lobes were frozen in liquid nitrogen at a temperature of -196°C, then transferred into the cryostat chamber and sections 15 μm thick were prepared at a temperature of -16°C. To obtain the needed topographic parts of the brain, the pieces were so oriented as to take sections in the frontal plane. The prepared sections were pasted on chemically pure slides and dried at room temperature (+20°C). In view of the great importance of enzymatic systems that regulate energy metabolism in the organism, we determined the activity of the key enzymes of the citric acid cycle--succinate dehydrogenase (SDH)--1.3.99.1 (by the method of E. Pearse), pentose cycle--glucose-6-phosphate dehydrogenase (G-6-PDH)--1.1.1.49 (also by the method of E. Pearse), tissular respiration--NAD-H_2-tetrazolium oxyreductase (NAD-H)--1.6.2.1 (by the method of M. Berston), glycolysis--hexokinase (HK)--2.7.1.1 (by the method of Sigel and Pette). The sections were incubated for 40 min at a temperature of +37°C in the dark. After this, they were eluated in saline, fixed in 10% neutral formalin in 0.9% sodium chloride, washed in 20° ethyl alcohol and imbedded in polyvinyl alcohol. The activity of the tested enzymes was graded by the method of V. I. Kolodin and O. K. Kuznetsov, according to which the following scores were used: (-)--no reaction (±)--traces of reaction (+)--mild, (++) --moderate, (+++)--marked and (++++)--very high activity.

The cortex and region of associative and commissural pathways of the rabbit brain were submitted to histochemical examination.* We analyzed the perikaryon of neurons referable mainly to the 3d-4th layers and astroglial elements of the cerebral cortex and oligodendroglial elements in white matter. The results are listed in the table.

Activity of enzymes in different types of brain cells

Type of glia examined	SDH		G-6-PDH		NAD-H		HK	
	control	experim.	control	experim.	control	experim.	control	experim.
Neuronal body	+ ±	+	++++	++	++++	++	+	±
Astroglia	+ ±	±	+++	++	+++	++	+	±
Oligodendroglia	+	±	++++	++±	+++	++	+	±

Very high G-6-PDH and NAD-H activity, moderate SDH activity and mild HK activity were demonstrated in neuronal bodies of the 3d-4th layers of the cortex. Analysis of the reaction for G-6-PDH revealed that intensive formation of diformasan granules was inherent not only in neuronal cortex, but astroglial and oligodendroglial cells. The very compact arrangement of dark-blue granules in the cytoplasm and processes was indicative of increased deposition of diformasan. Moreover, diformasan granules were demonstrable in the fibers of white matter conductive pathways. In some cases, fragments of the choroid plexus of cerebral ventricles, ependymal and subependymal layers were demonstrable in brain sections, in which G-6-PDH activity was also rather high. NAD-H activity was usually high in the perikaryon of cortical cells, moderate or marked in astroglia and oligodendroglia. Deposition of diformasan granules similar in shape and color was also noted in nerve fibers. With demonstration of SDH, moderate deposition of diformasan granules was found both in the bodies of nerve cells referable to the pyramidal and ganglionic layers, and cytoplasm of astrocytes of the protoplasm type. The granules were fine and brownish-lilac in color. The SDH reaction was relatively low in oligodendroglial elements.

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*I. Sh. Galeyev and N. V. Zgoda participated in these studies.

HK activity was rather low in the cellular elements examined, with the exception of capillary endothelium, where the activity of this key enzyme of glycolysis was marked in three cases.

Consequently, very high G-6-PDH activity and low HK activity are noted in the brain of control animals, which is indicative of prevalence of the phosphorylase pathway of replenishing the intracellular supply of glucose-6-phosphate and significant contribution of the pentose pathway to utilization of intermediates of carbohydrate metabolism. There was also high NAD-H and moderate SDH activity, on the basis of which we can conclude that there is prevalence of the NAD-H-dependent pathway of transport of reducing equivalents in the respiratory chain and some contribution of the succinate dehydrogenase system to this process.

According to intensity of formation of diformasan granules in the perikaryon of neurons of the 3d and 4th cortical layers in experimental animals, the tested reactions were arranged in the following descending order: NAD-H, G-6-PDH, SDH and HK. The level of NAD-H activity was characterized by deposition of dark blue, fine, similar granules of diformasan, which were uniformly distributed in the bodies of cortical neurons. In demonstration of G-6-PDH activity, deposition of diformasan was maximal in pyramidal cells and least marked in the molecular, external granular and polymorphous cortical layers. SDH activity was low and occasionally moderate in all cortical layers; the diformasan granules were small, similar in shape and of a brownish lilac color. We also demonstrated traces of HK activity. Testing of NAD-H activity in astrocytic glia revealed moderate formation of diformasan granules. The reaction for G-6-PDH also revealed moderate activity; the astrocytes demonstrated very low activity in SDH and HK. Oligodendroglial elements along the course of nerve fibers presented moderate G-6-PDH and NAD-H activity. There were similar, fine, dark blue diformasan granules in the oligodendrocyte cytoplasm, but they were virtually undemonstrable in the dendrites of neurons. A trace reaction was inherent in cytoplasm of oligodendrocytes in demonstration of SDH and HK.

Consequently, there was significant decrease in G-6-PDH activity in the brain of experimental rabbits, which is indicative of depression of the pentose pathway of utilization of glucose-6-phosphate in a hypomagnetic environment. In turn, this could be due to either accumulation of intermediators of the pentose pathway, which inhibit G-6-PDH activity, or decreased need for synthesis of NAD-H and riboses. We also demonstrated a moderate decline in activity of NAD-H and SDH, which is indicative of overall decrease in metabolic activity of cellular elements of the cerebral cortex of experimental rabbits, since the level of activity of these enzymes reflects, to a significant extent, the intensity of reactions in Krebs' cycle.

Thus, a hypogeomagnetic field is a biologically active factor, which causes a decrease in activity of key enzymes of metabolism in cellular elements of the rabbit's cerebral cortex.

We do not have data at the present time as to the stage of mammalian development at which it is affected by a hypogeomagnetic field, the consequences of longer exposure of animals to hypogeomagnetic conditions, the effect this factor would have on offspring, the genetic system, which biological processes and functional systems are the most sensitive to a hypogeomagnetic field, etc. Further complex studies of this problem are needed to identify the mechanisms of biological effects of a hypogeomagnetic field and to work out means of protection, since our findings indicate that it is desirable to consider the adverse effect of a hypogeomagnetic field in medical monitoring of working conditions for personnel, in particular pregnant women, working in special installations.

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